

Isolation of Non-Cytopathic (NCP) VSV Mutants

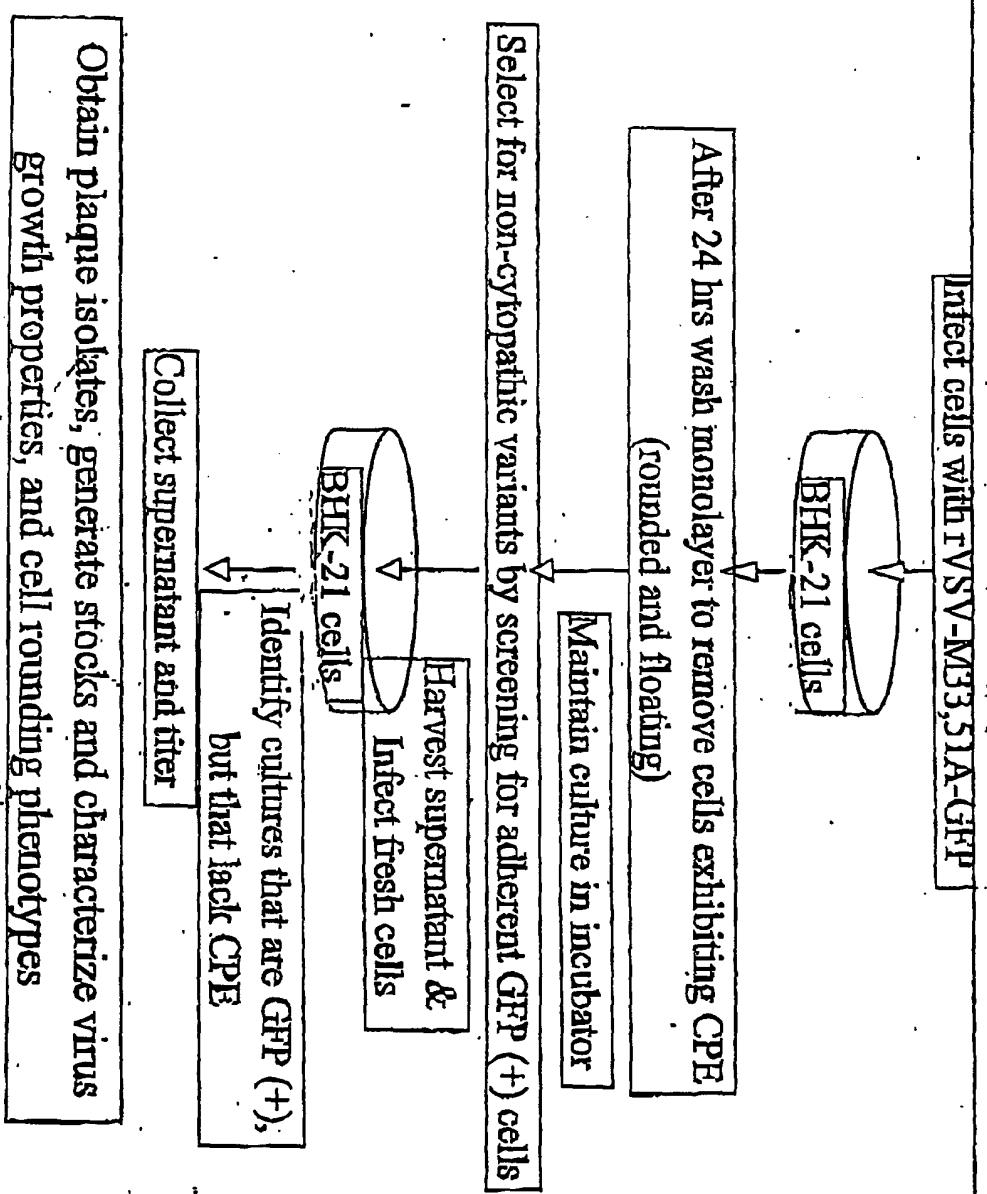
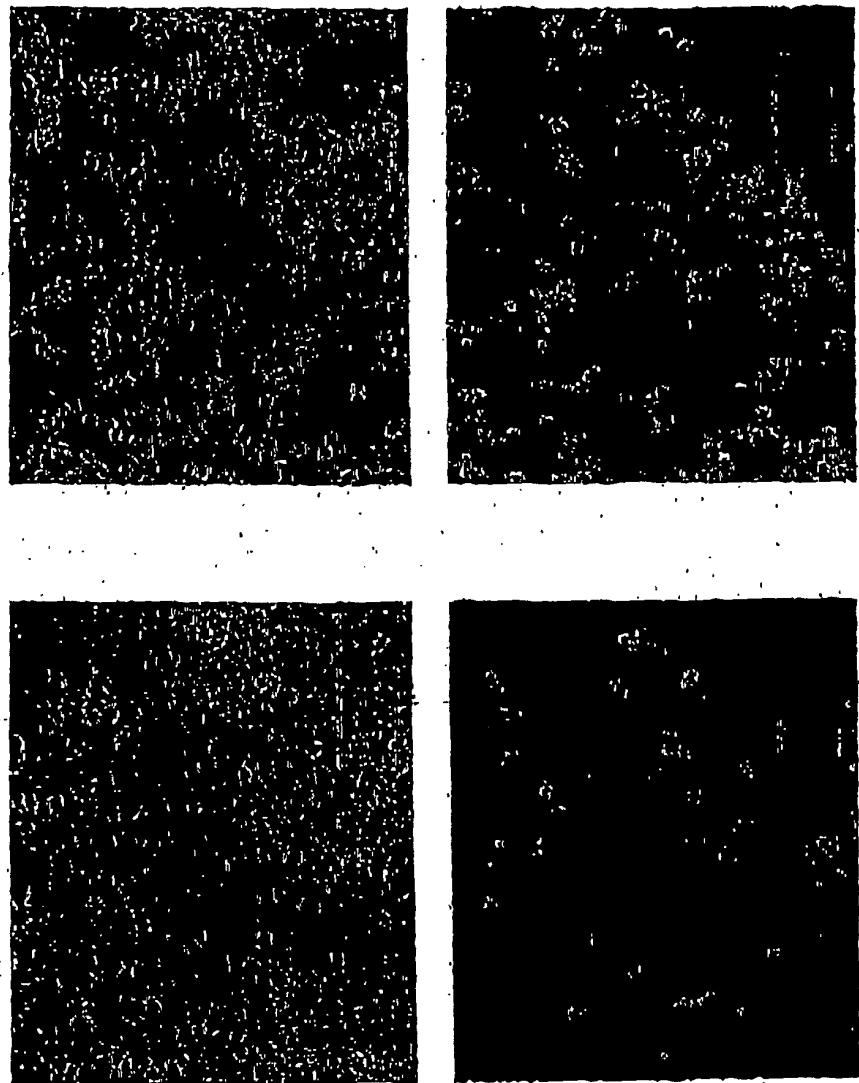


Figure 1

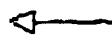
Cell Rounding Phenotypes of M Protein Mutants of VSV



Isolation and Sequencing of NCP-M

Protein Mutant cDNAs

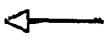
Purify NCP virus from culture supernatants of plaque purified NCP isolates



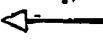
Isolate genomic RNA and perform RT-PCR using M gene specific primers



Subclone cDNAs into plasmid pBS-SK+



Sequence individual clones and identify mutations present in M_{NCP} cDNAs

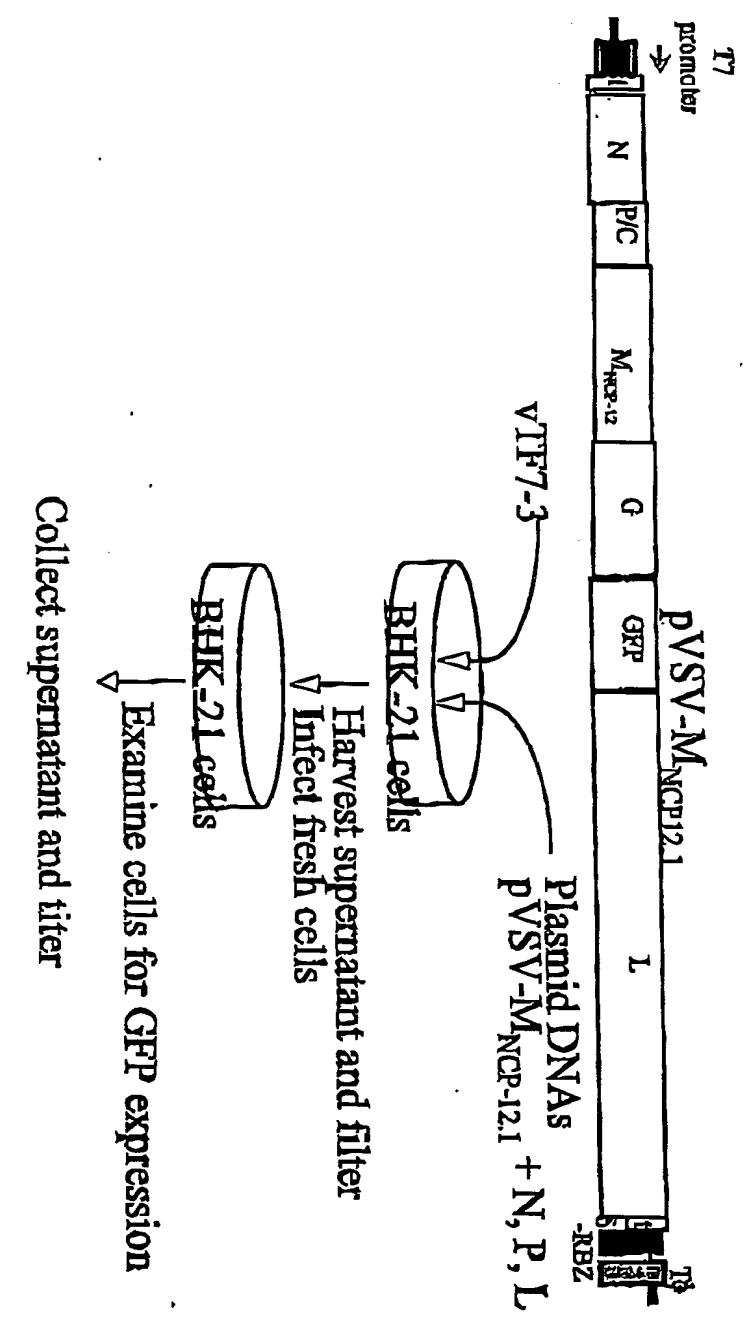


NCP mutations are M33A; MS1A; T133A; & S226G
Designated M_{NCP12.1}

Subclone M_{NCP12.1} into pVSV-FL(+)2

Figure 3

Recovery of rVSV-M_{NCP12.1}



Obtain plaque isolates, generate stocks and characterize growth properties, and cell round

Figure 4

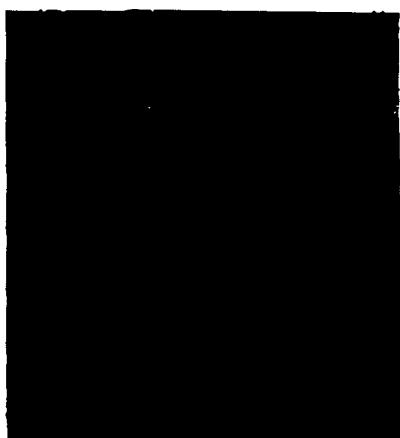
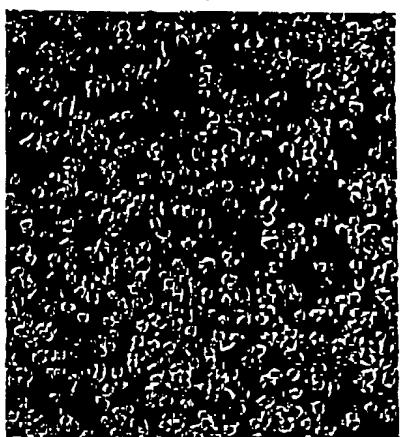
rVSV/M_{NCP12.1} is Defective in Cell Rounding

Phase Contrast

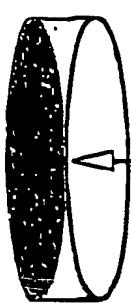
Anti-VSV N protein MAB

Infect BHK-21 cells

wtrVSV



Examine @ 12 hrs p.i.



rVSV
NCP-12.1

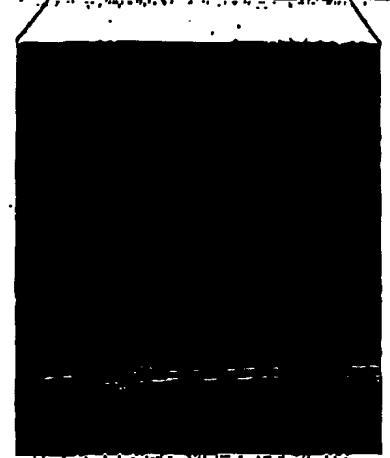
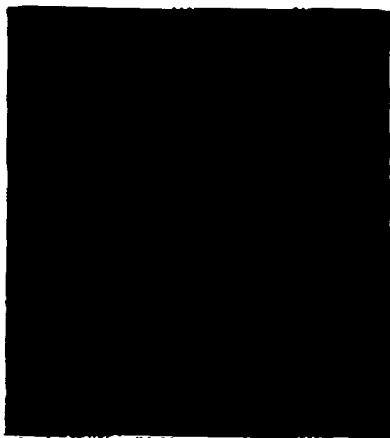


Figure 5

Transient Expression of M_{NCP-12.1}

wt M

NCP-12.1

NCP-12.1

mock

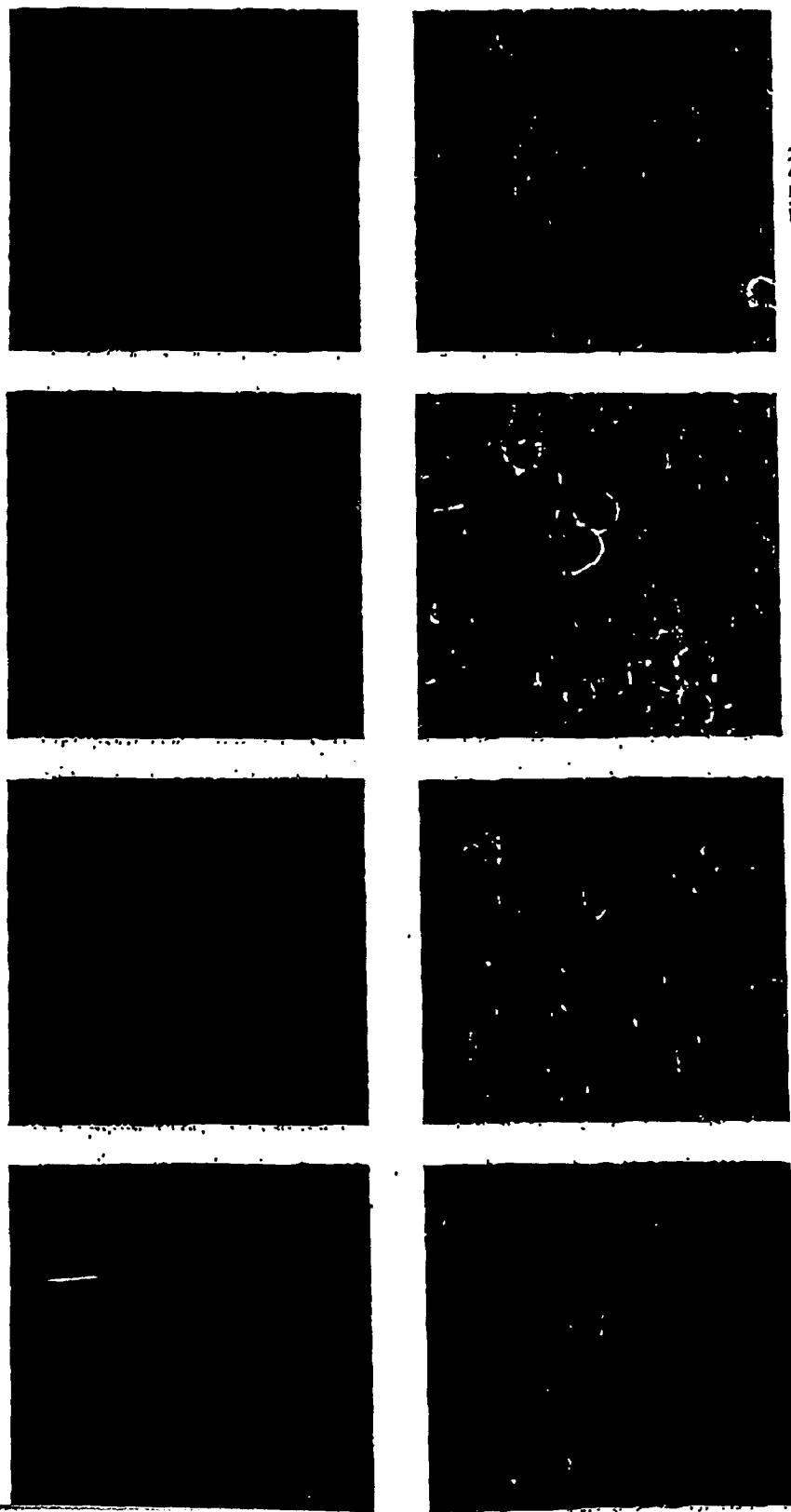


Figure 6

rVSV/M_{NCP-12.1} Infection of Different Cell Types

BHK @ 24 hr CV-1 @ 24 hr Hela @ 12 hr Vero @ 24 hr

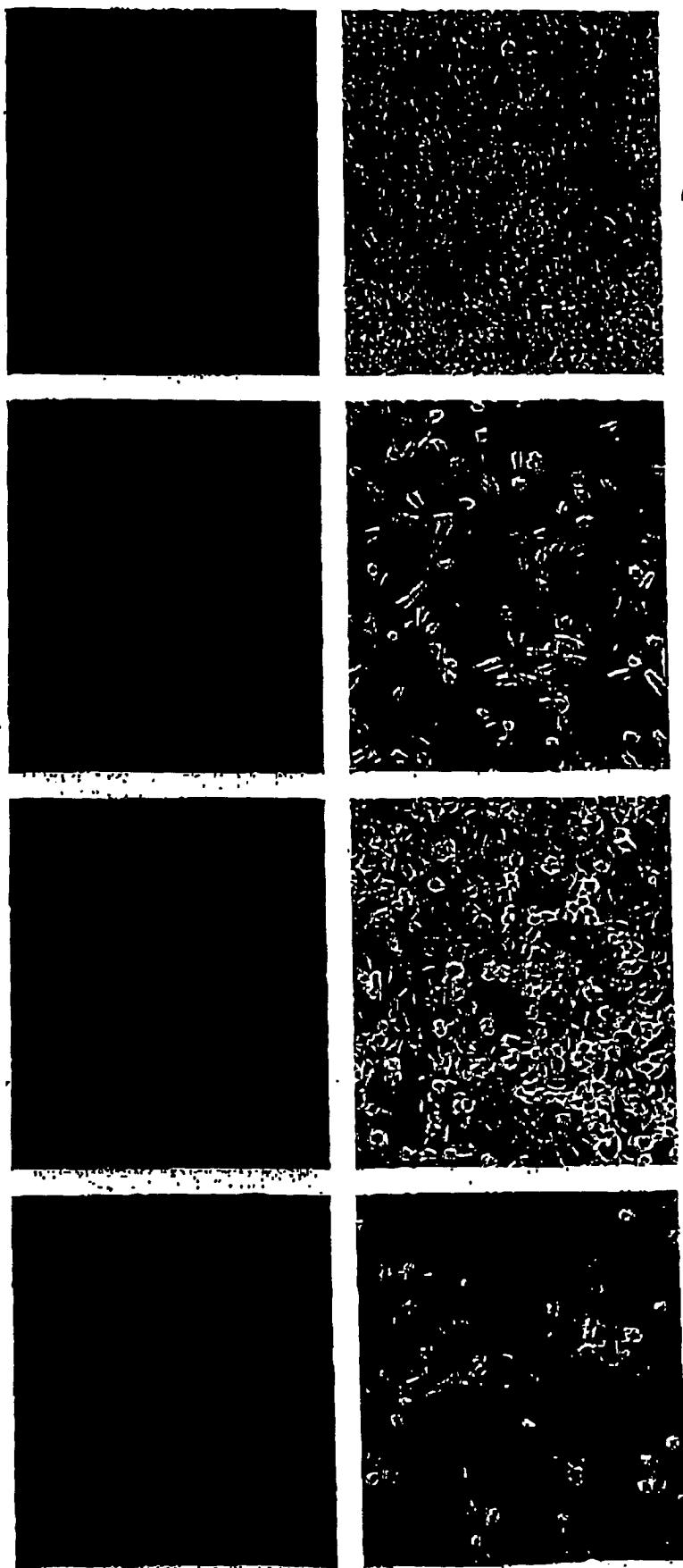


Figure 7

Use of M_{NCP-12.1} to Recover ΔM-VSV

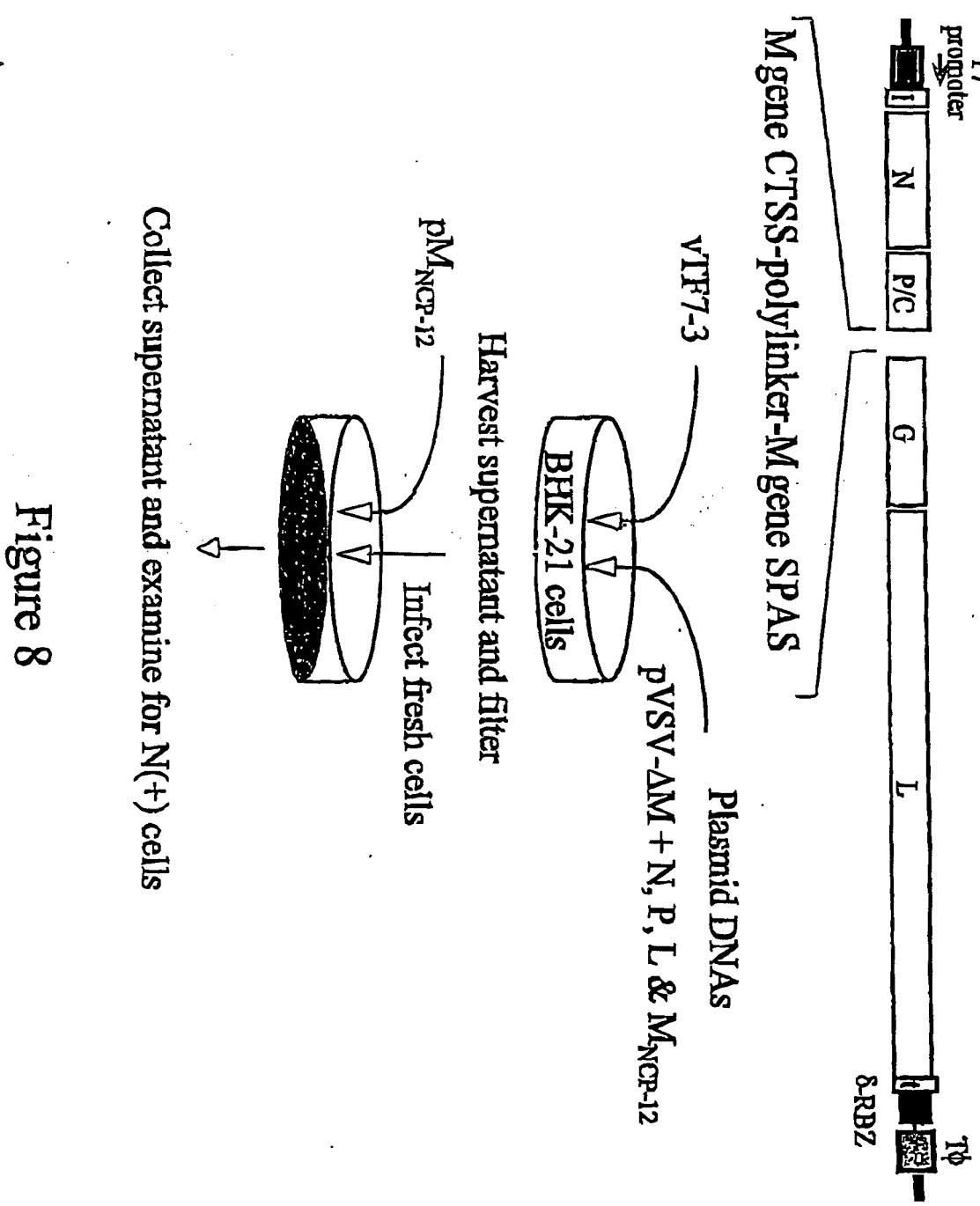


Figure 8

Recovery of rVSV- Δ M

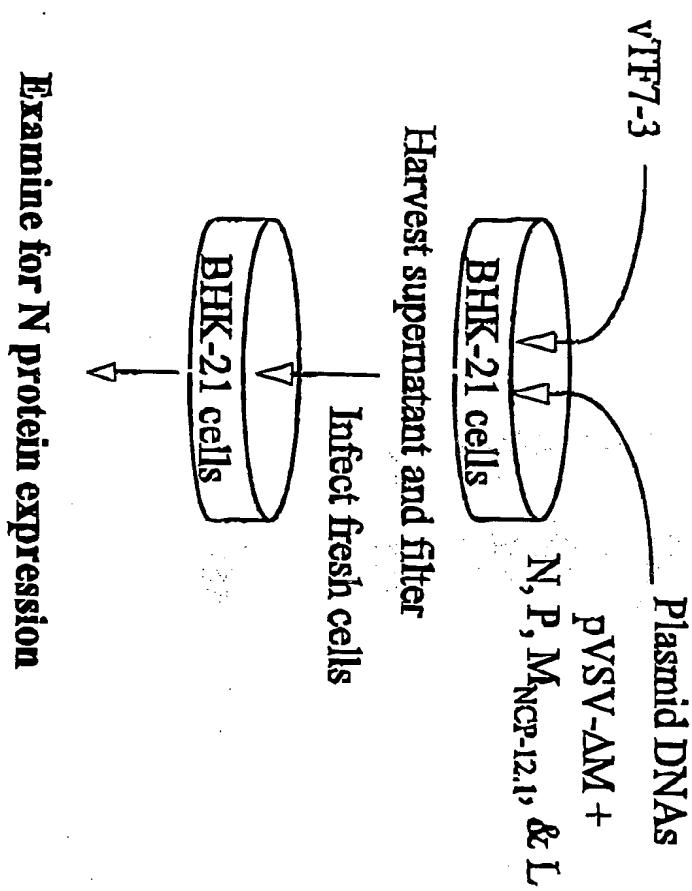
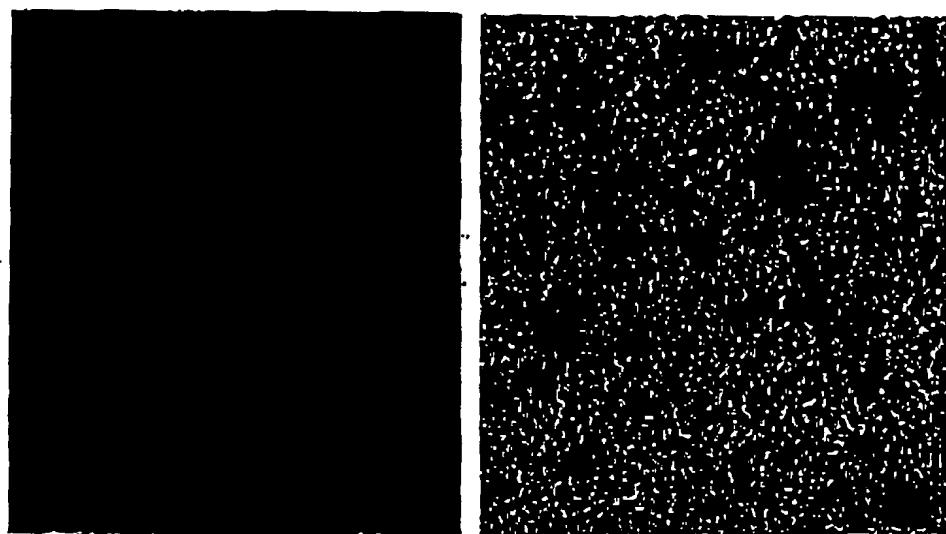


Figure 9



Sample #176, 3 Days Post-infection

Infection of islet Prep#176 at day 3 post infection

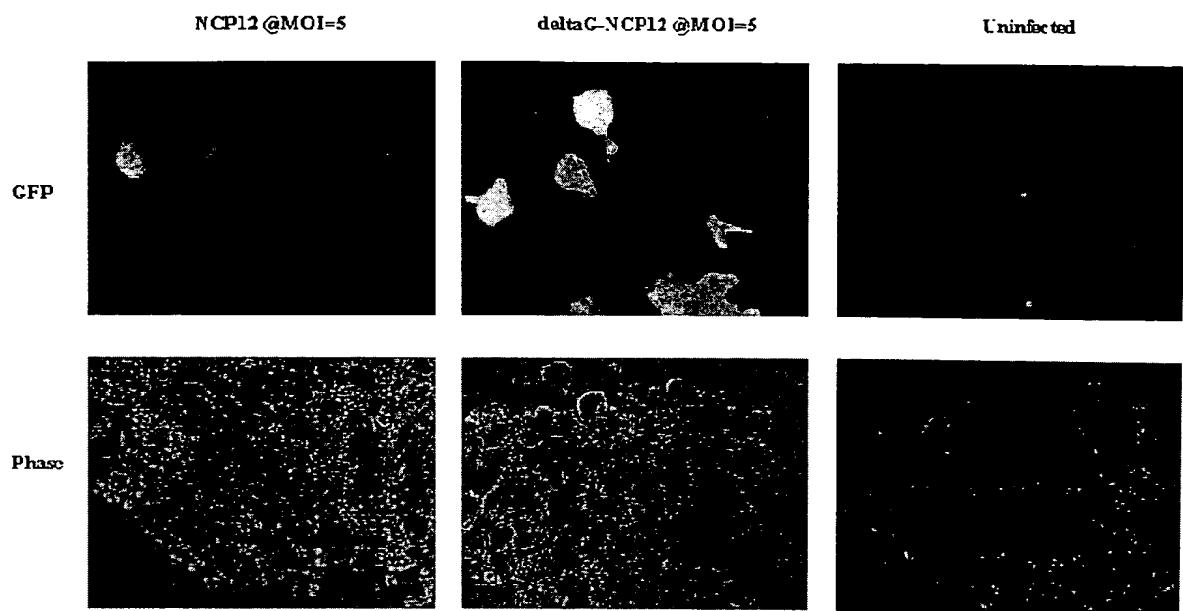


Figure 10

Sample #163, 3 Days Post-infection

Infection of islet Prep#163 at day 3 post infection

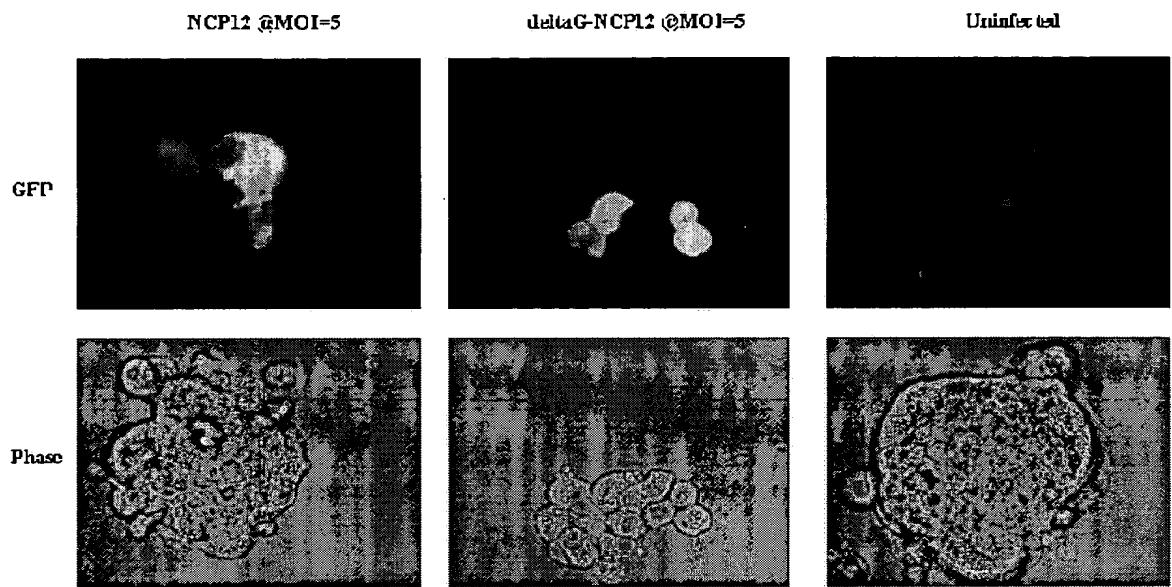


Figure 11

Sample #176, 3 Days Post-infection

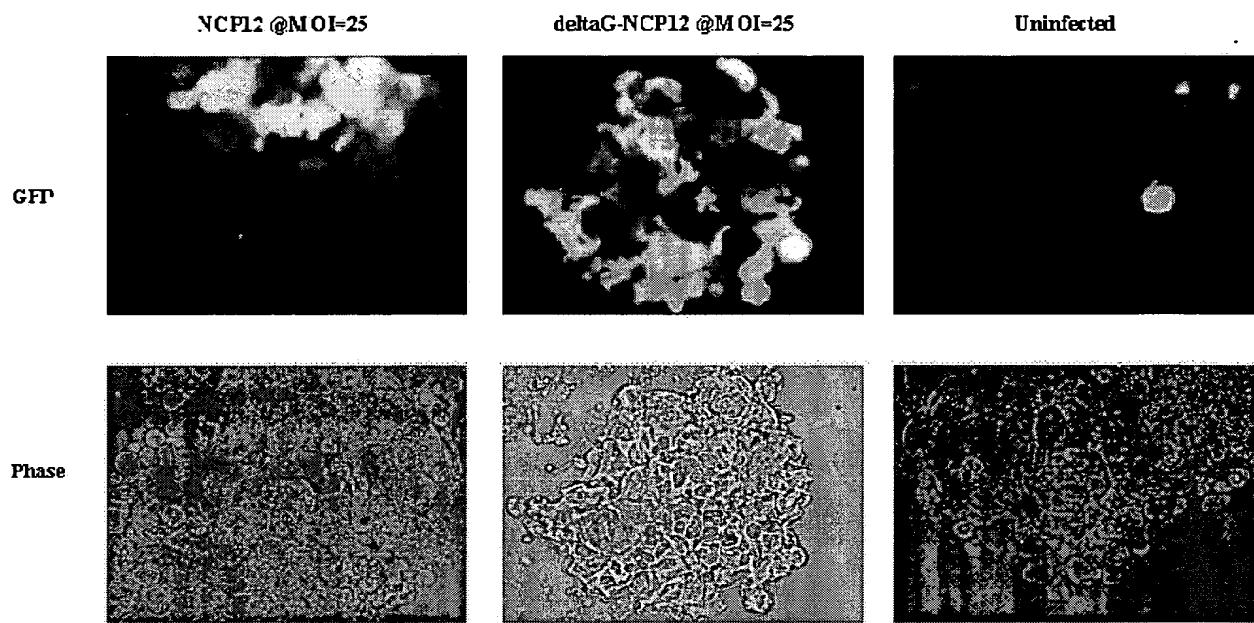


Figure 12

Sample #163, 3 Days Post-infection

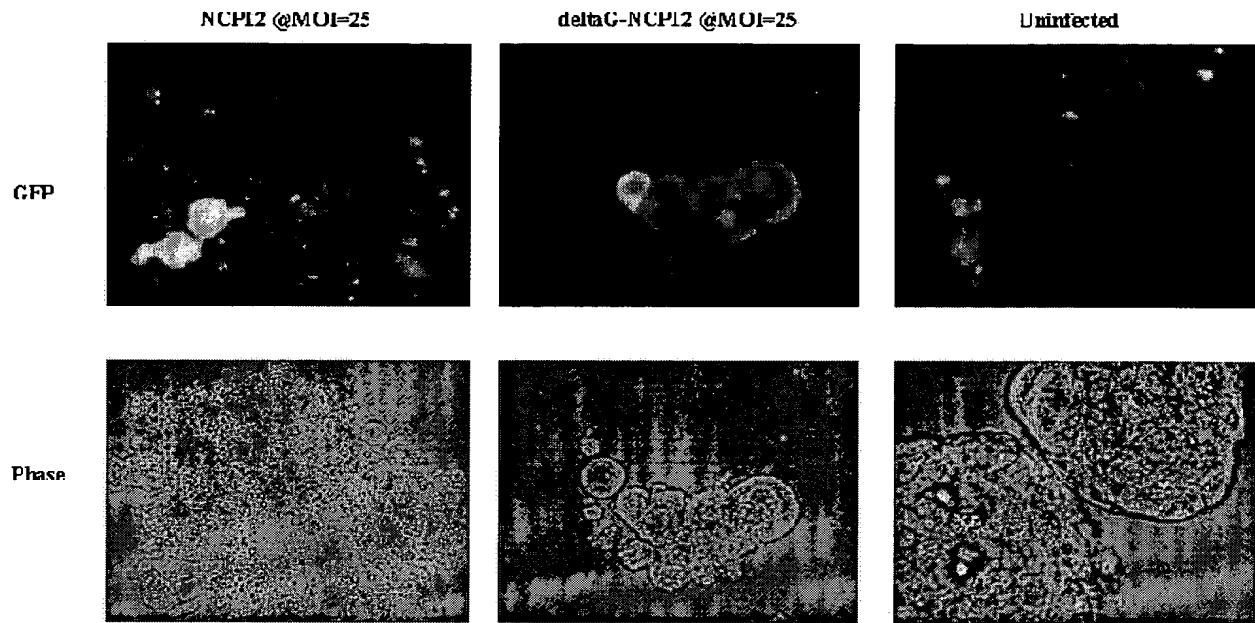


Figure 13

Sample #176, 8 Days Post-infection

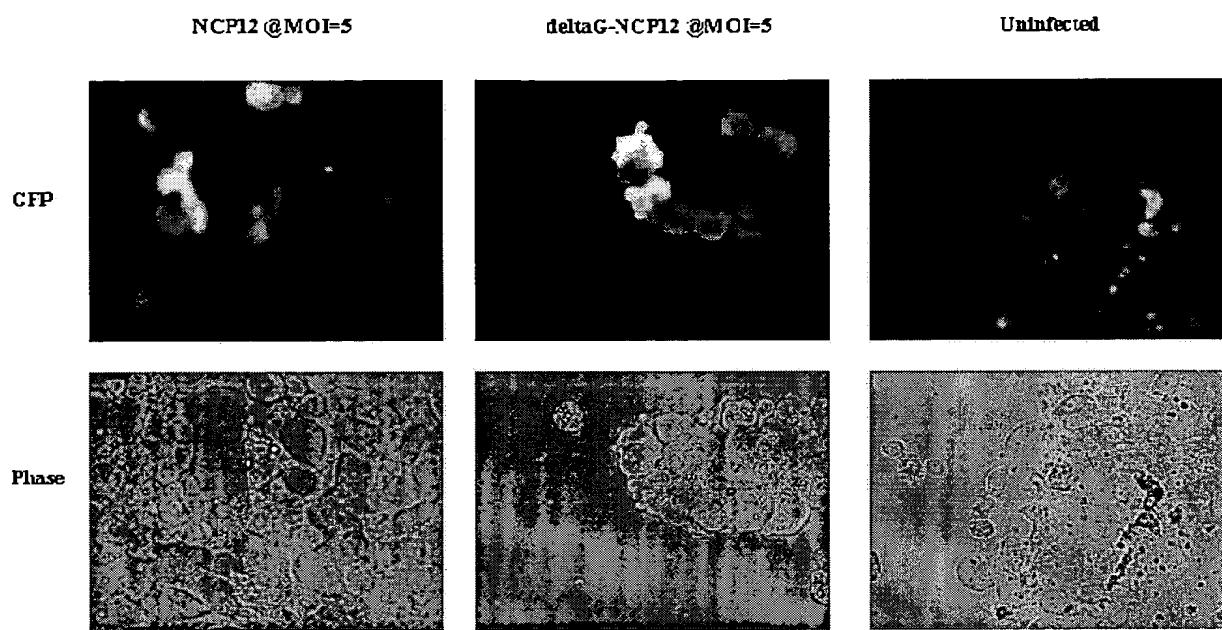


Figure 14

Sample #176, 8 Days Post-infection

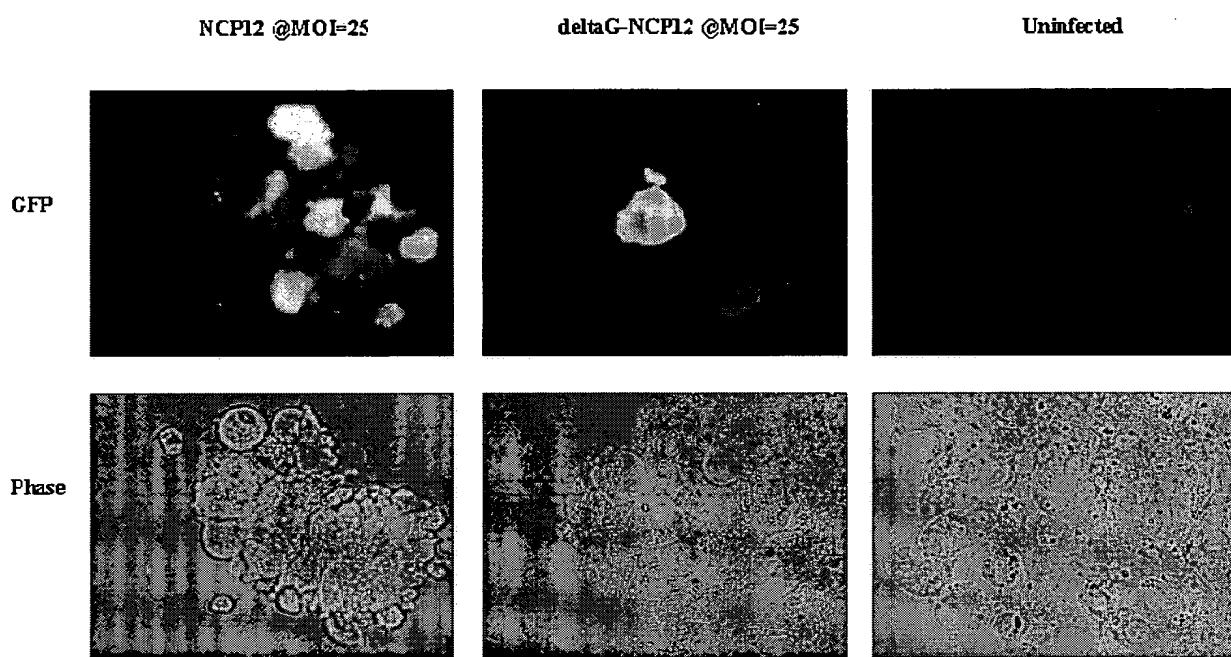


Figure 15

Sample #163, 8 Days Post-infection

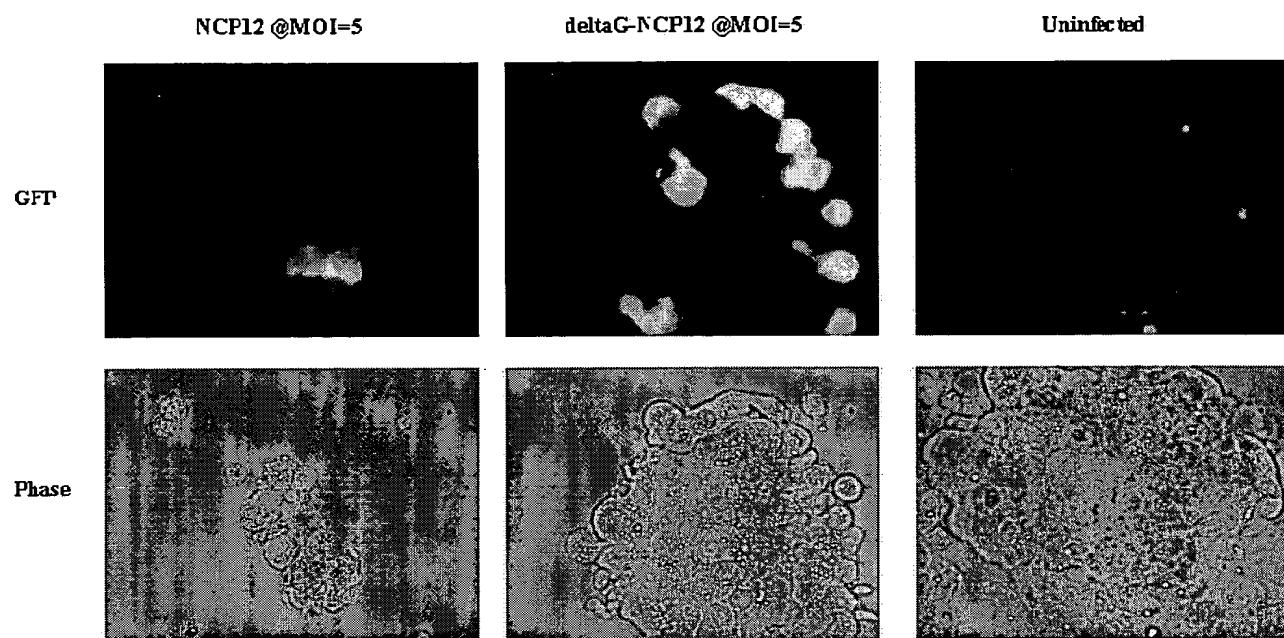


Figure 16

Sample #163, 8 Days Post-infection

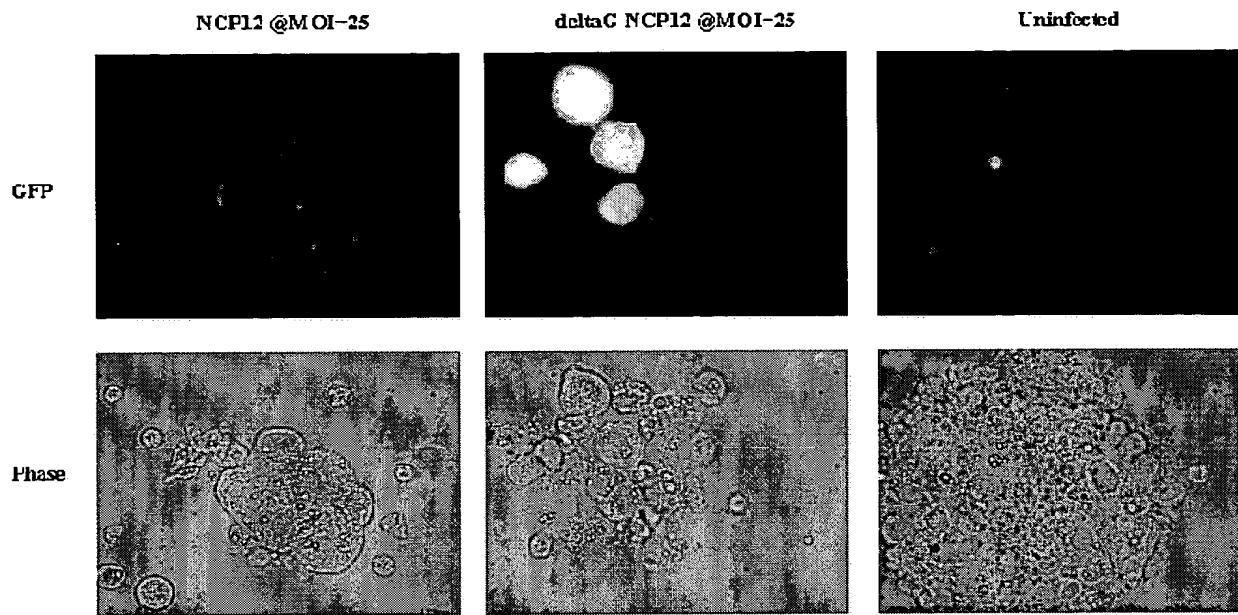


Figure 17

Sample #176, 3 Days Post-infection

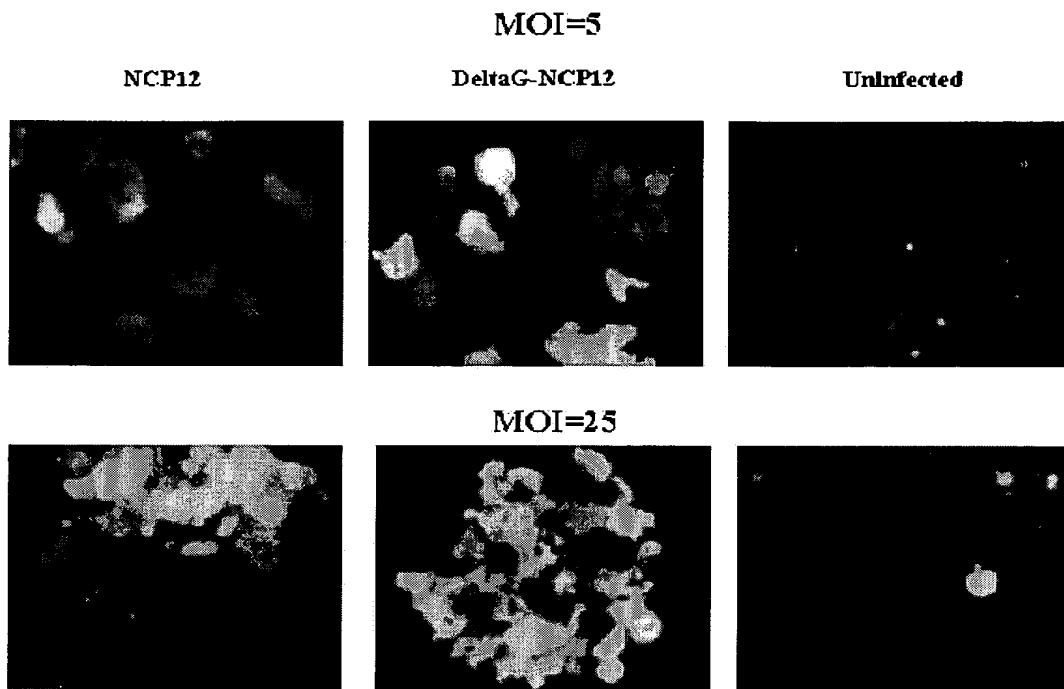


Figure 18

Sample #176, 8 Days Post-infection

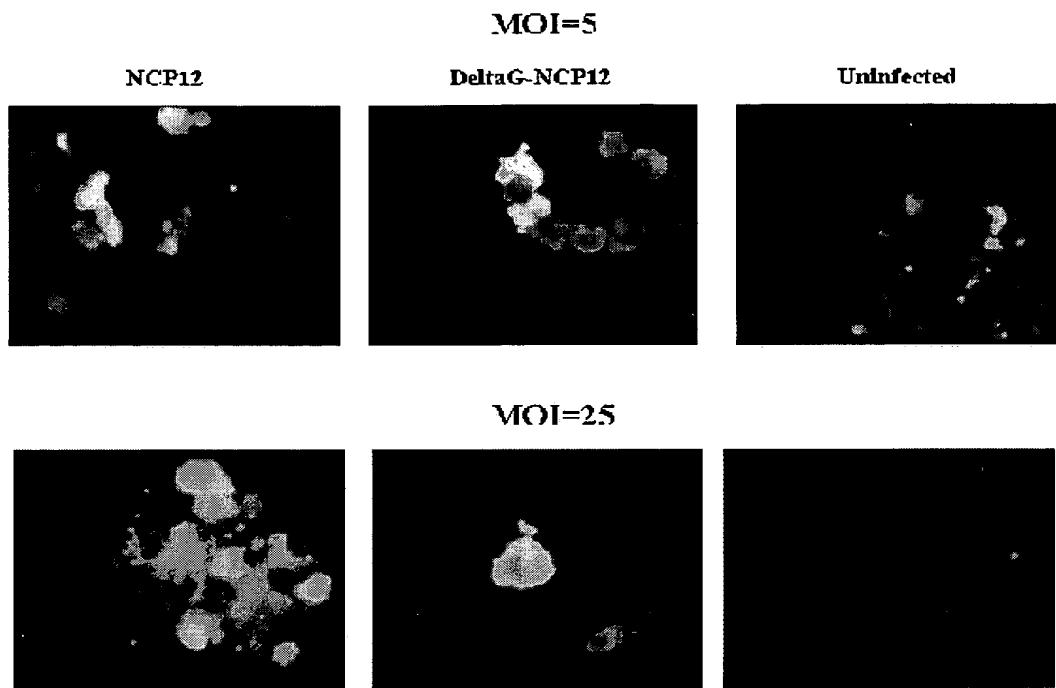


Figure 19

Figure 20.

Figure 21

Figure 22

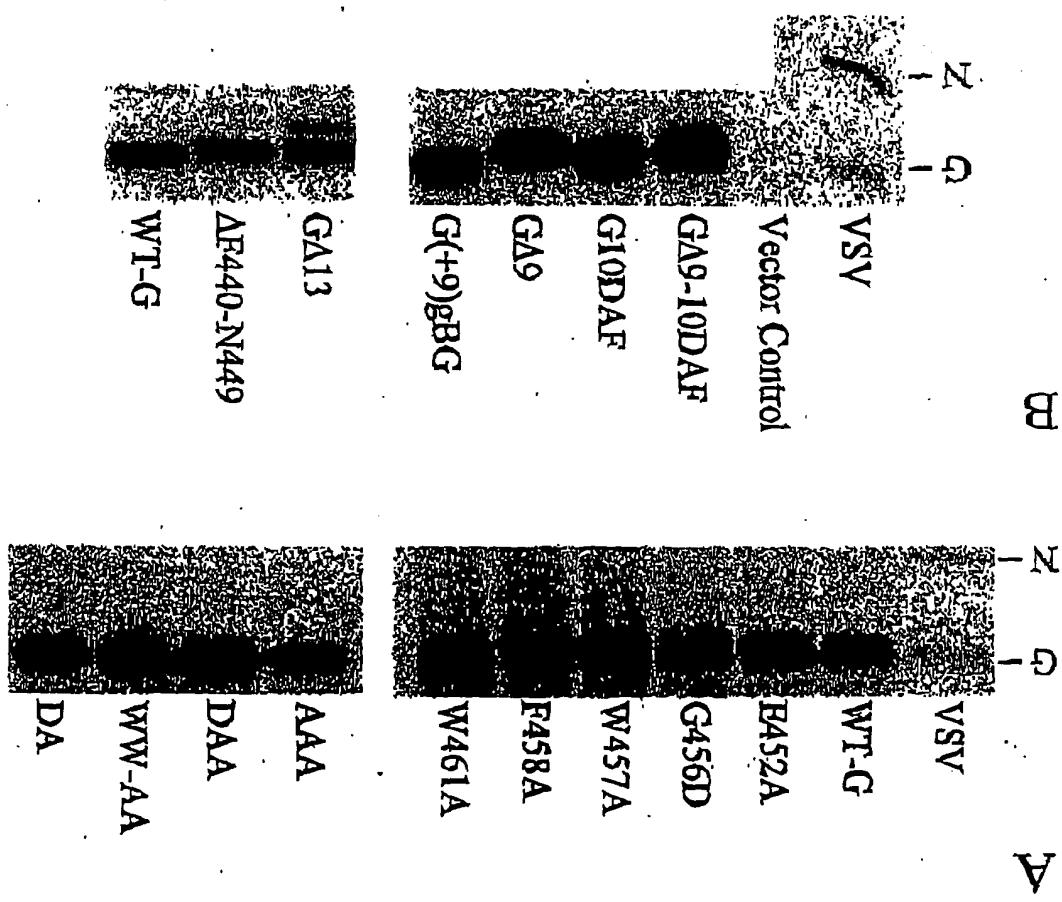


Figure 23

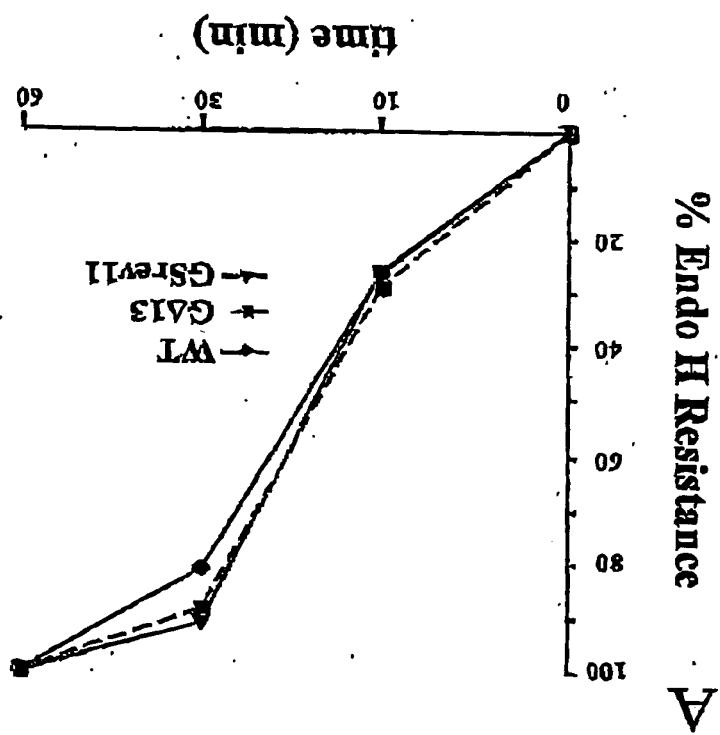
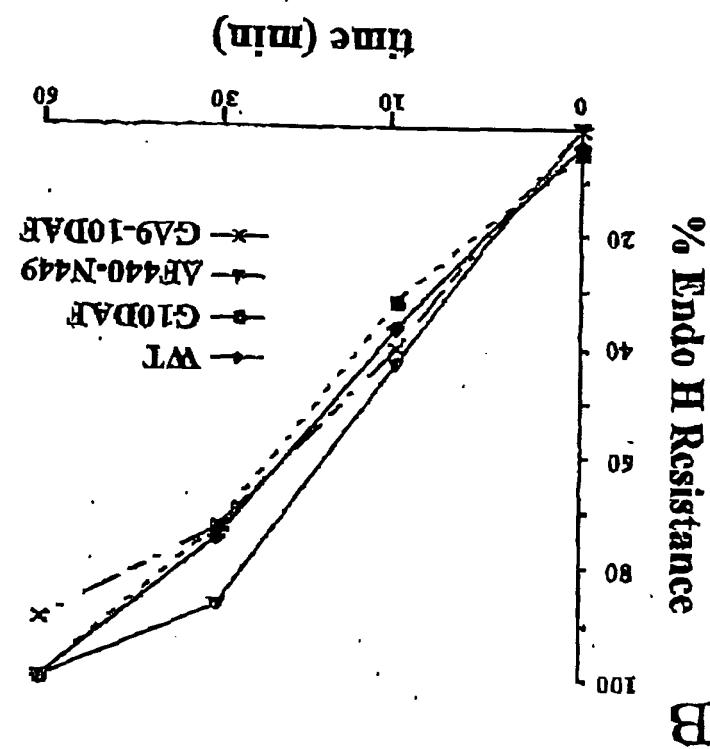
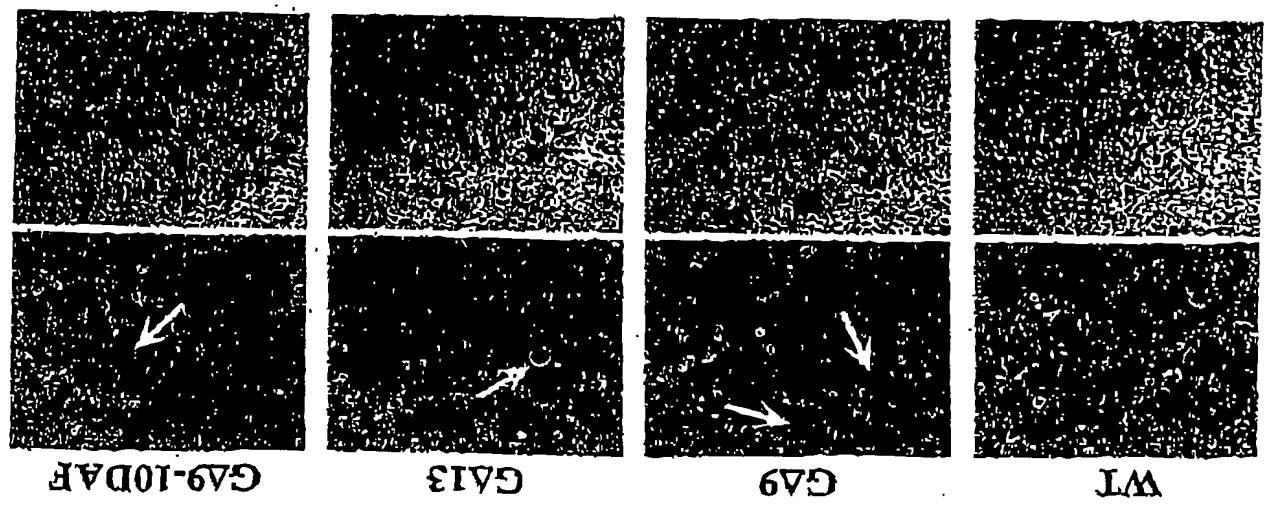
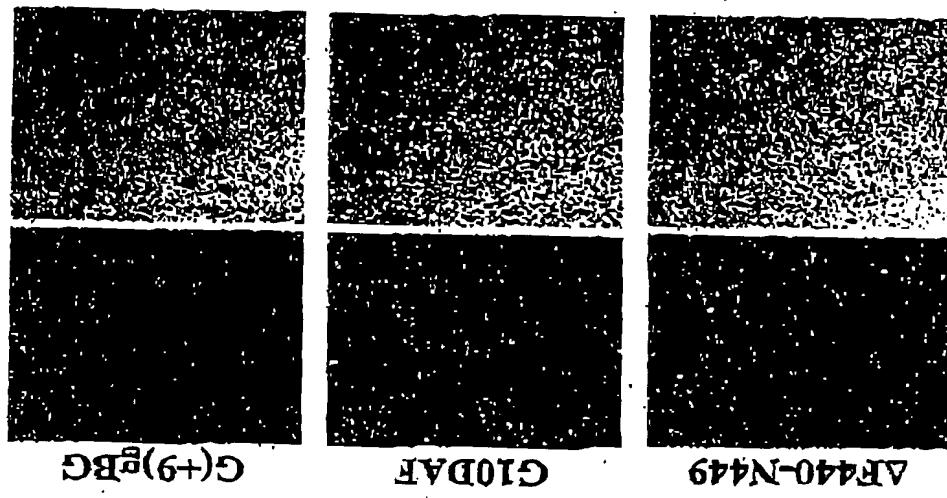


Figure 24



B

A

Figure 25

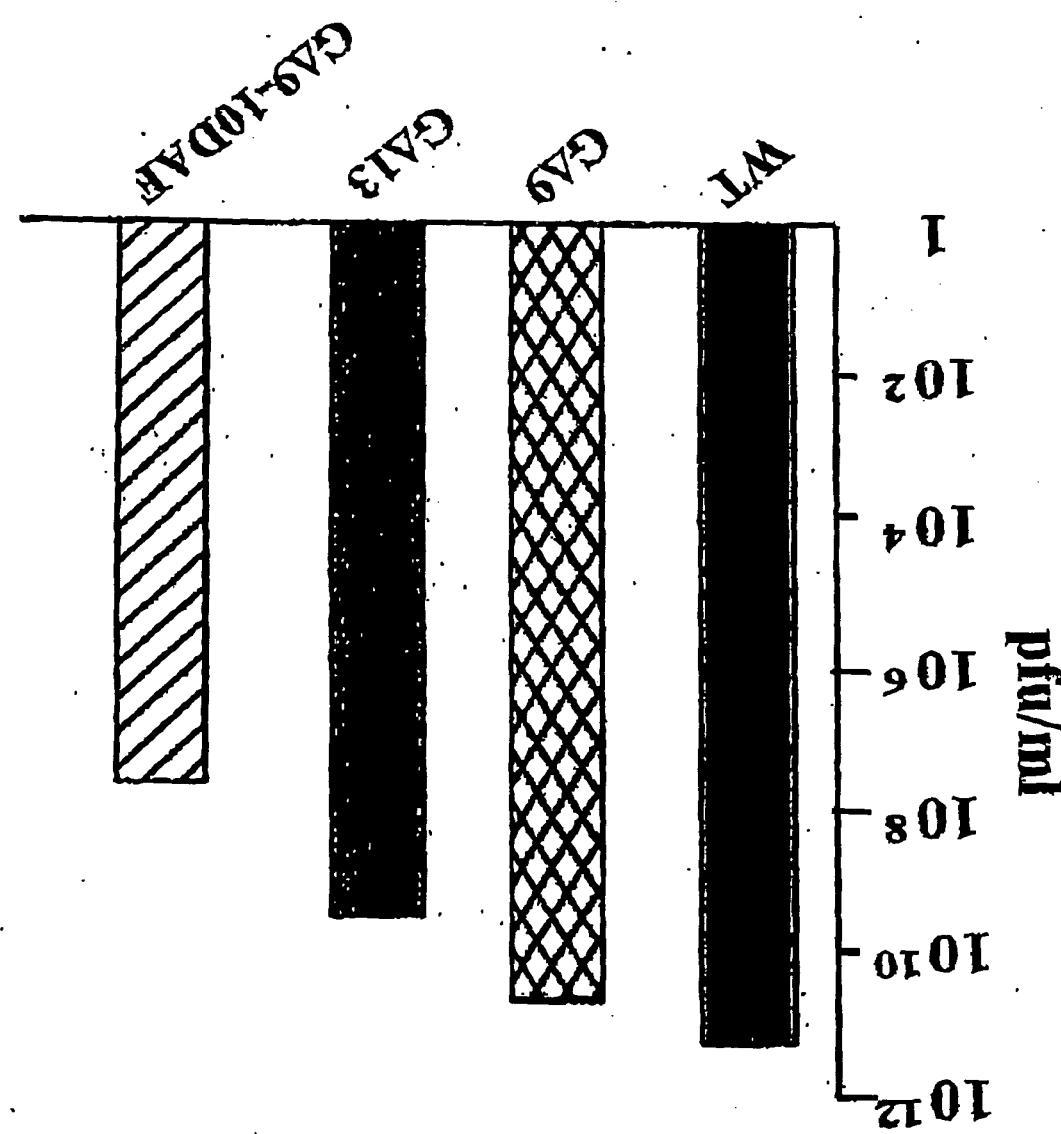


Figure 26

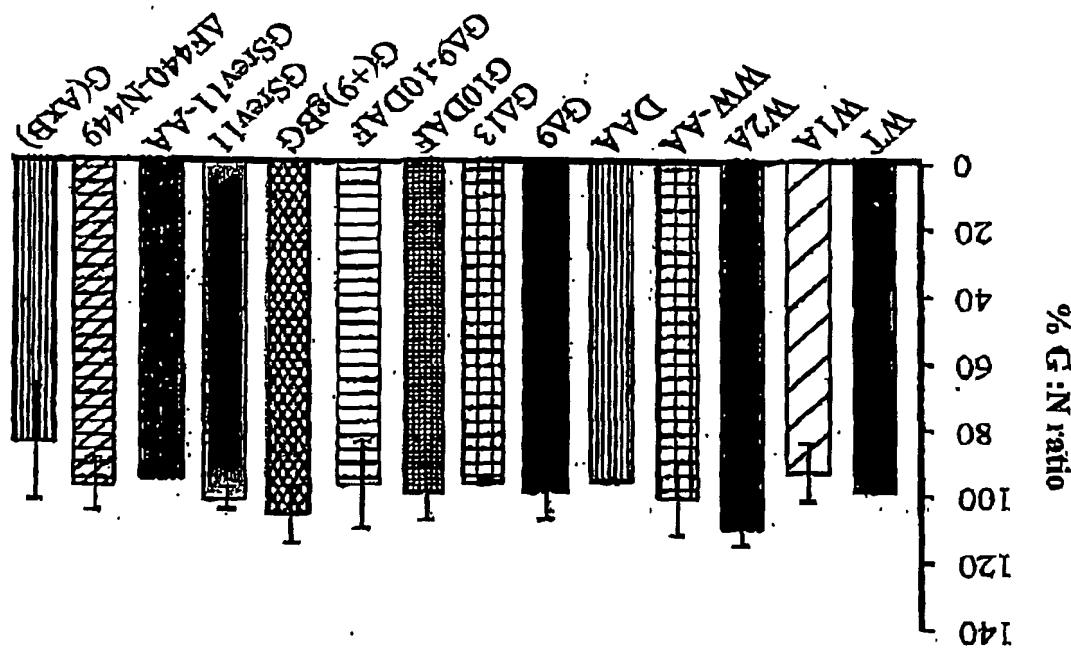


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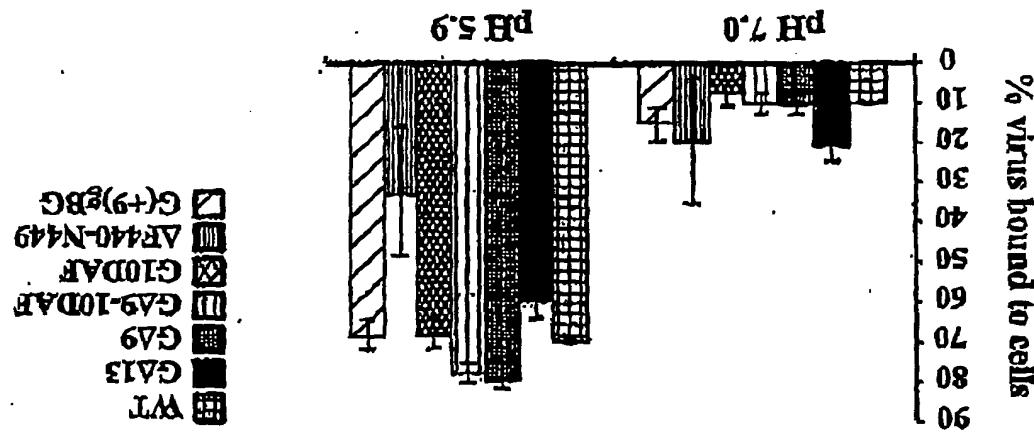


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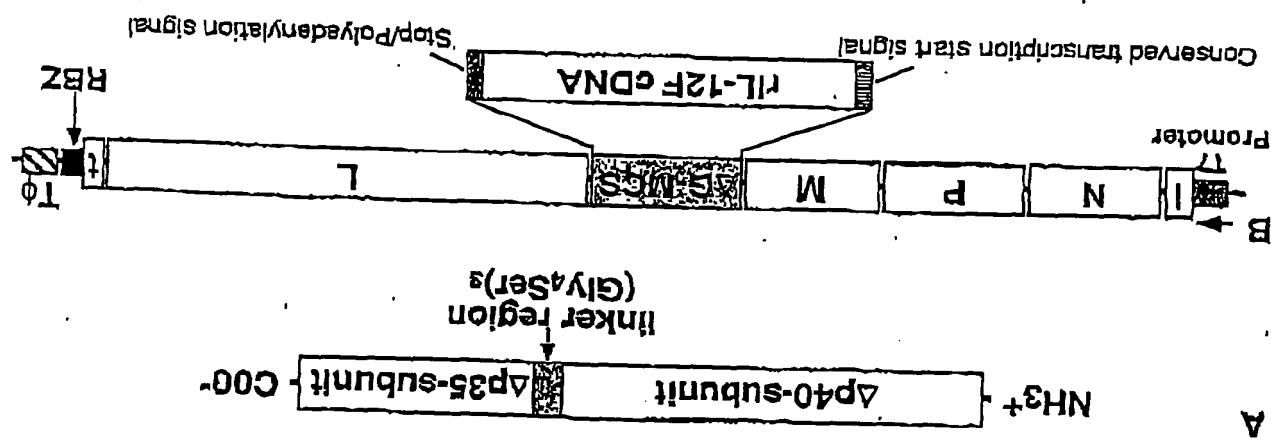
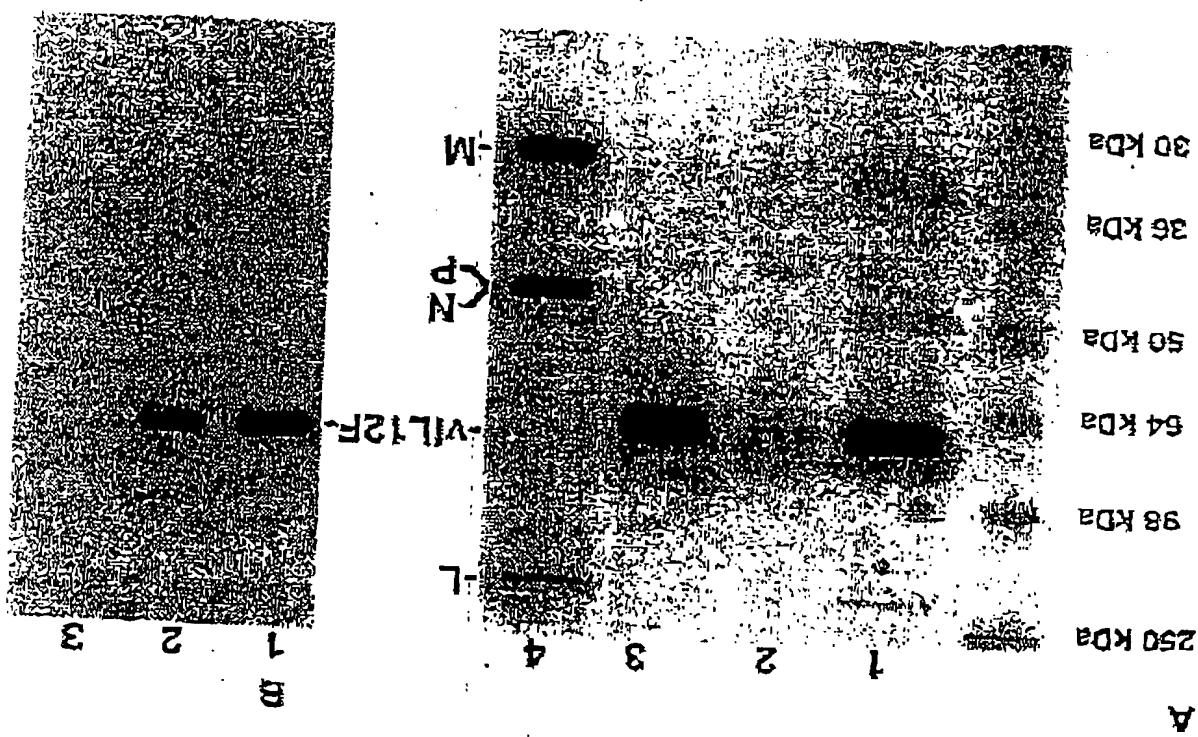


Figure 29



A

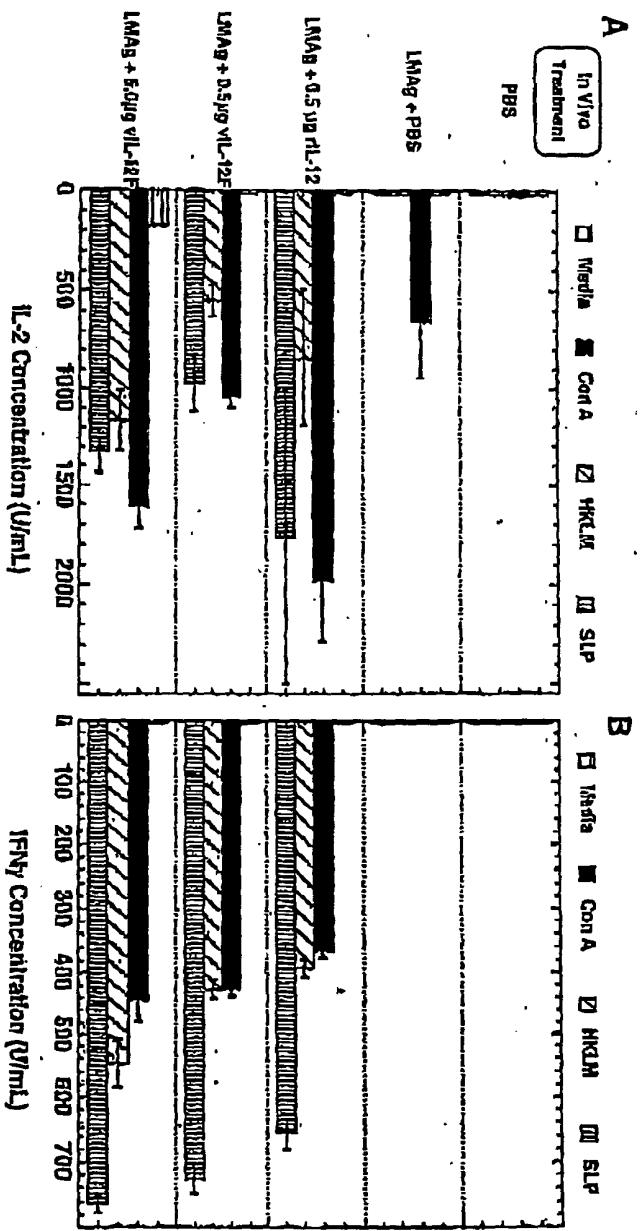
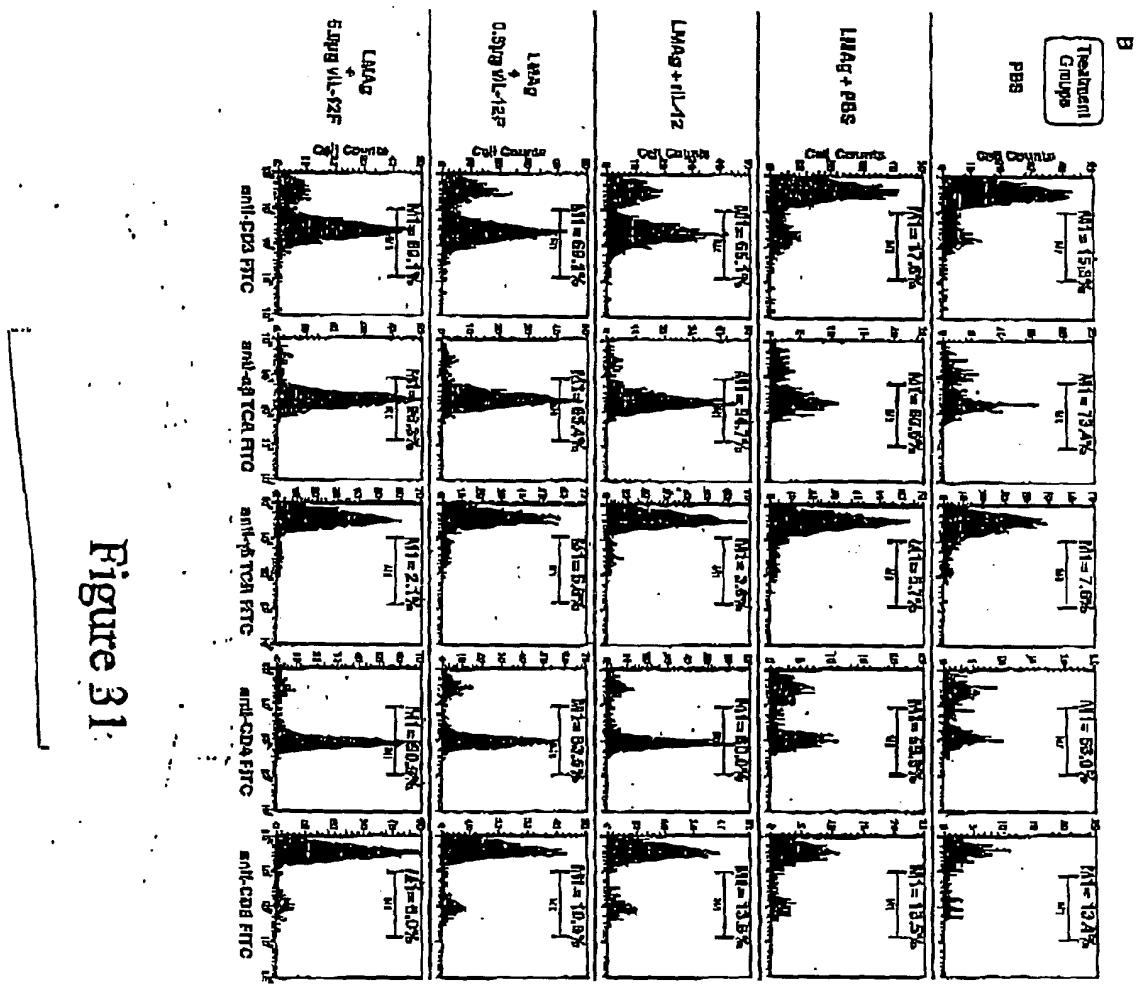


Figure 30

Figure 31:



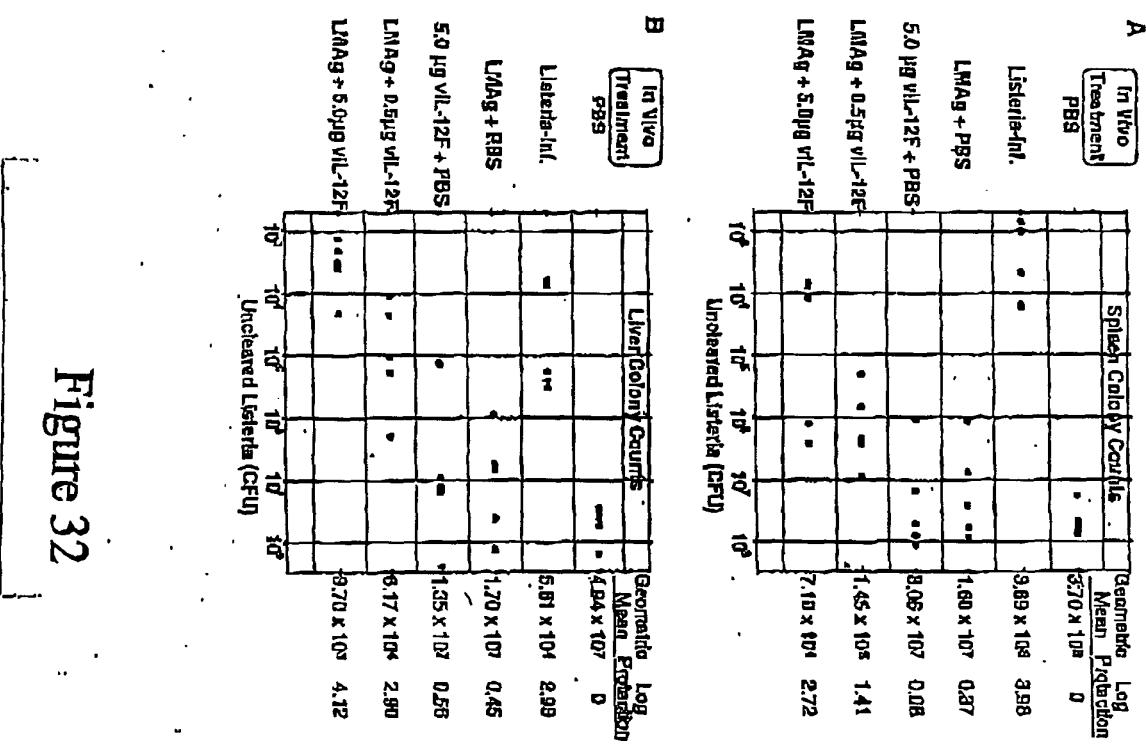


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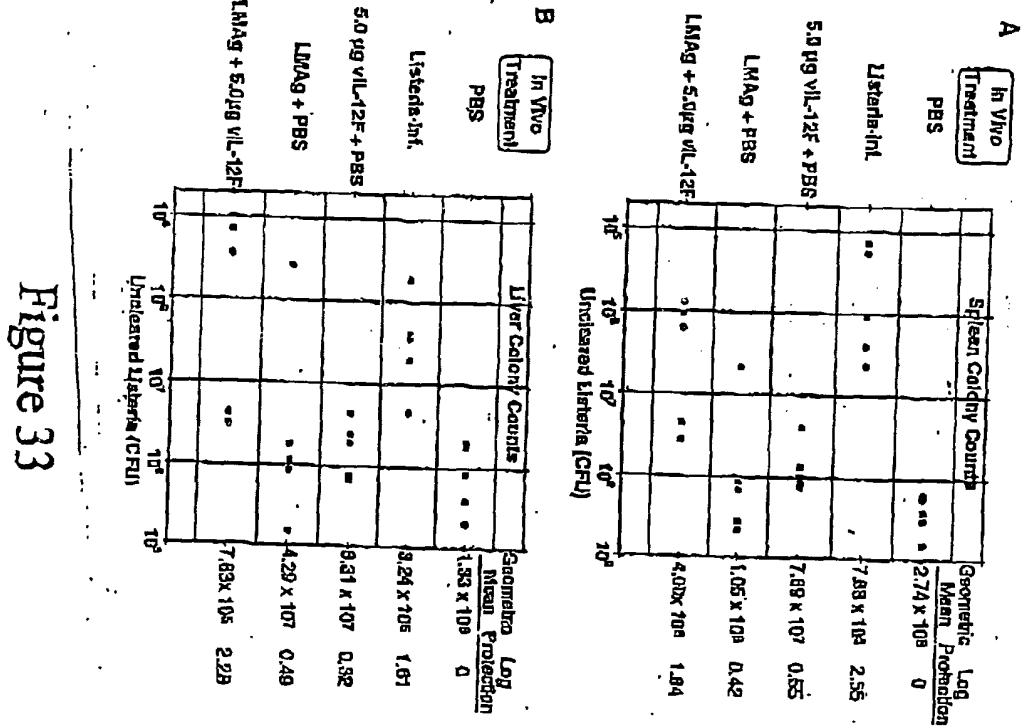
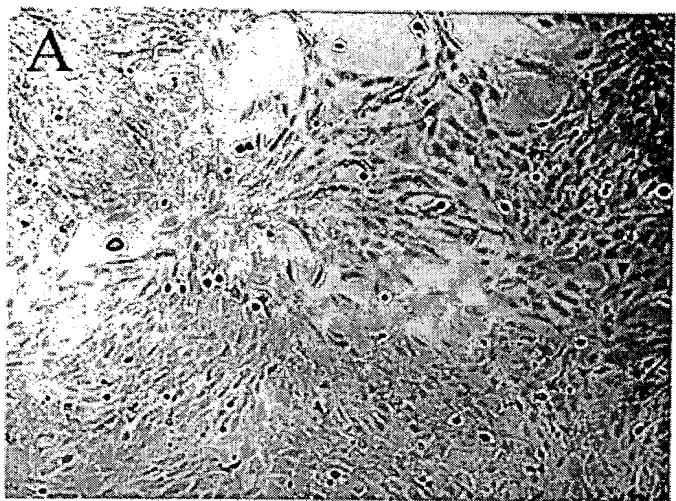
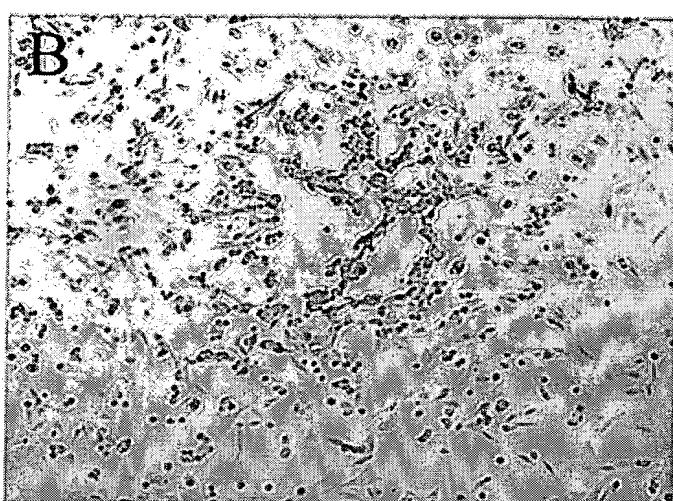


Figure 33

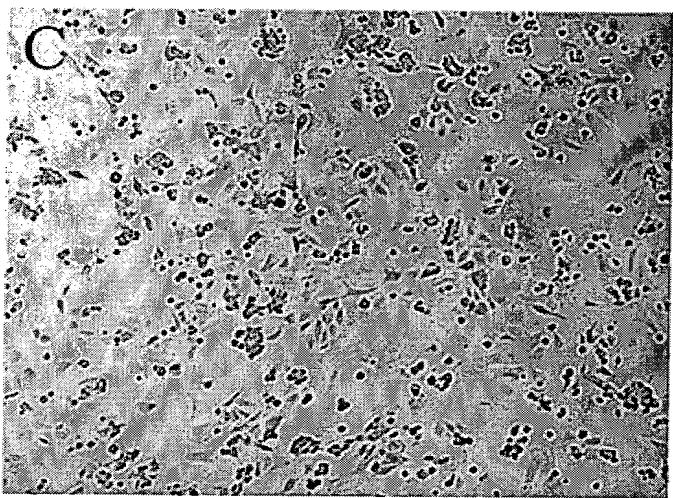
0 hr



10 hr



24 hr



48 hr

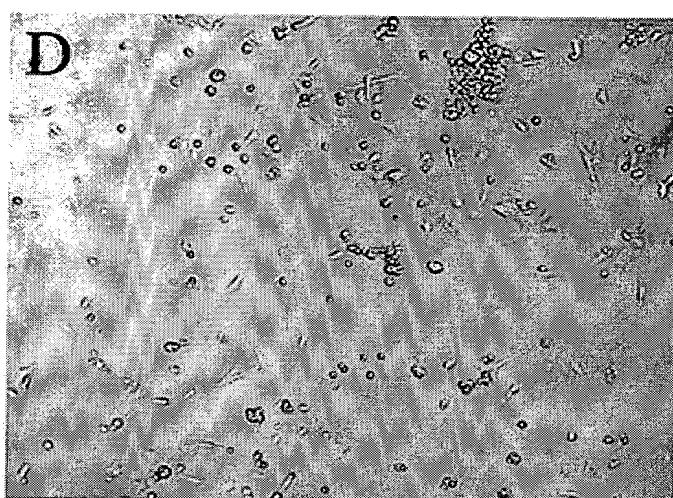


Figure 34

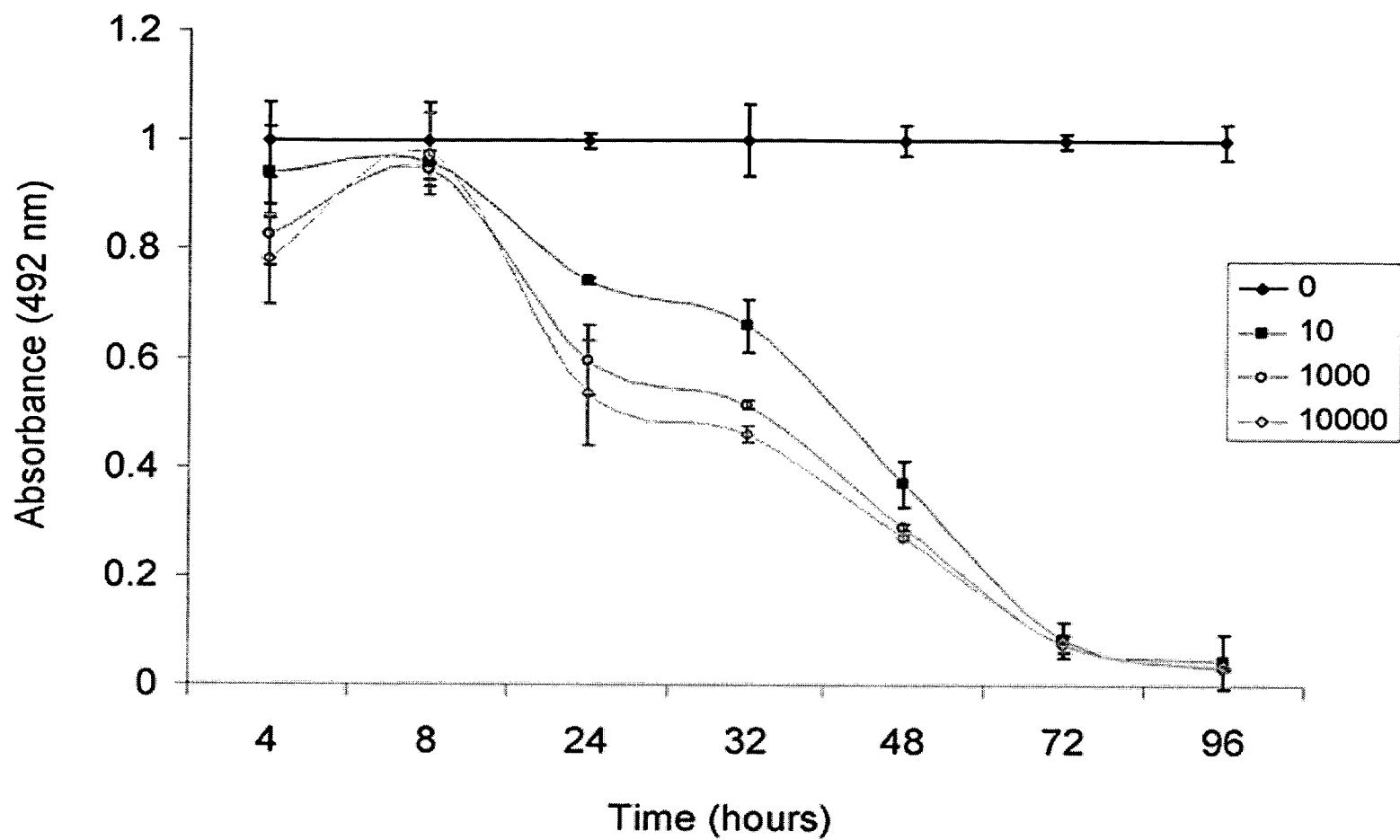
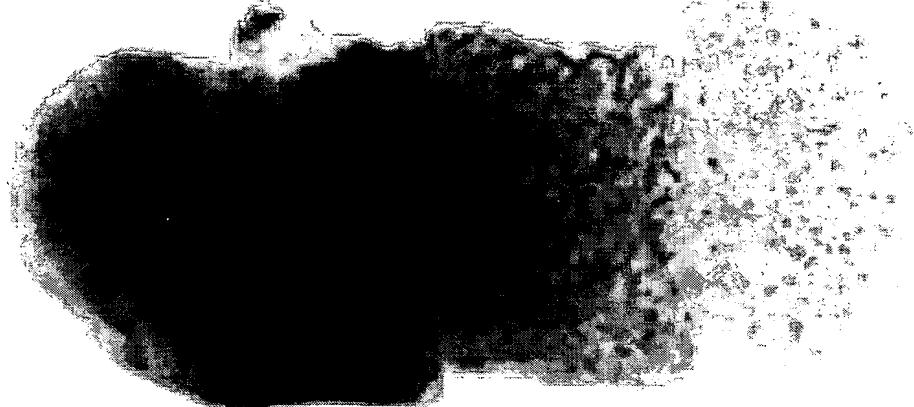


Figure 35

A

Substantia Nigra
(SN)

Striatum

Cortex
(Cx)

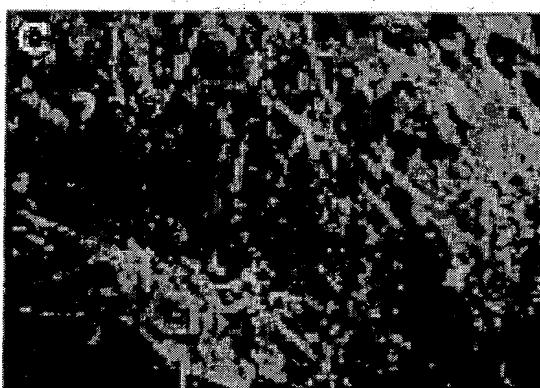
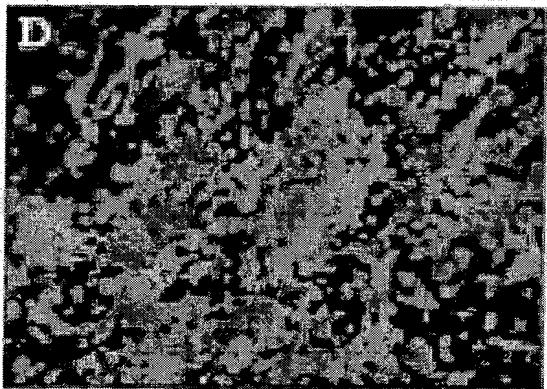
B**C****D****E**

Figure 36

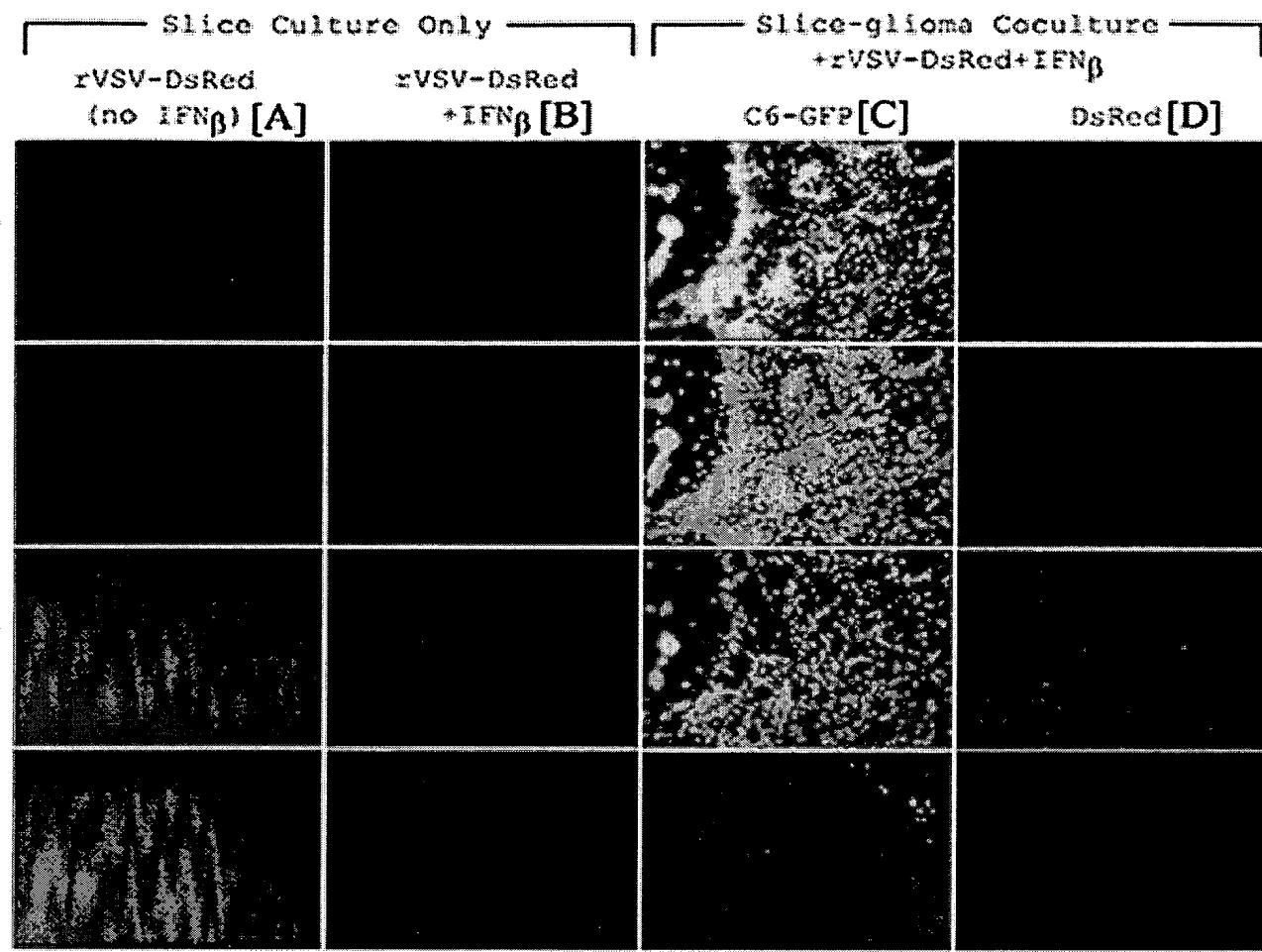


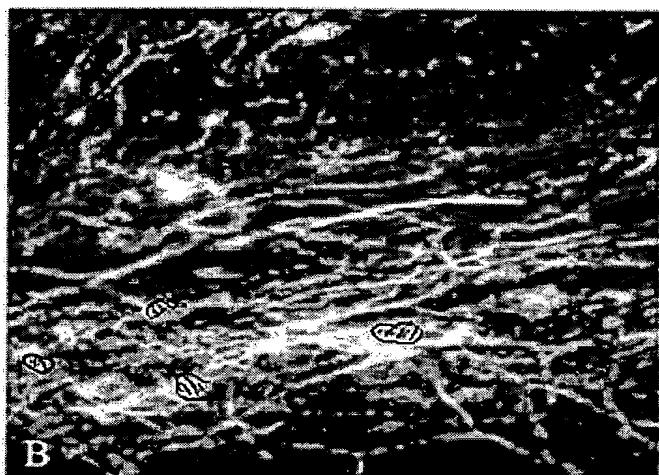
Figure 37

VSV-wt w/ no IFN

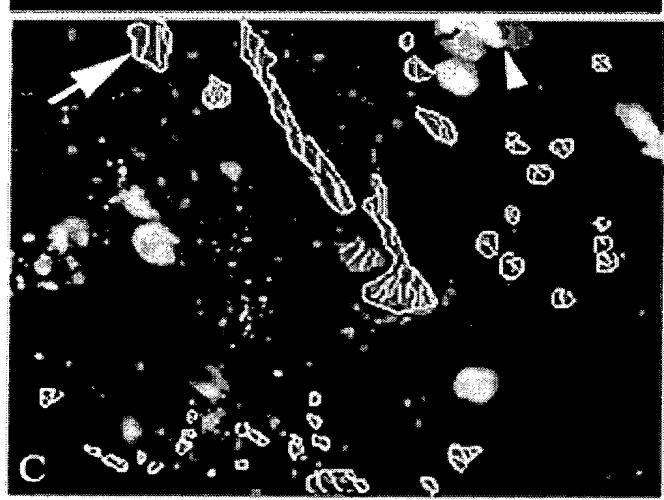


A

VSV-wt w/ 1,000u IFN

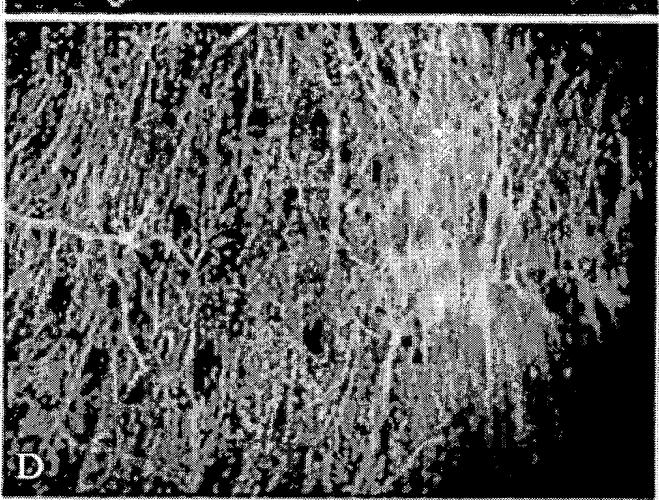


B



VSV-wt w/ 1,000u IFN on C6 co-cx

C



normal slice w/ IFN only

Figure 38

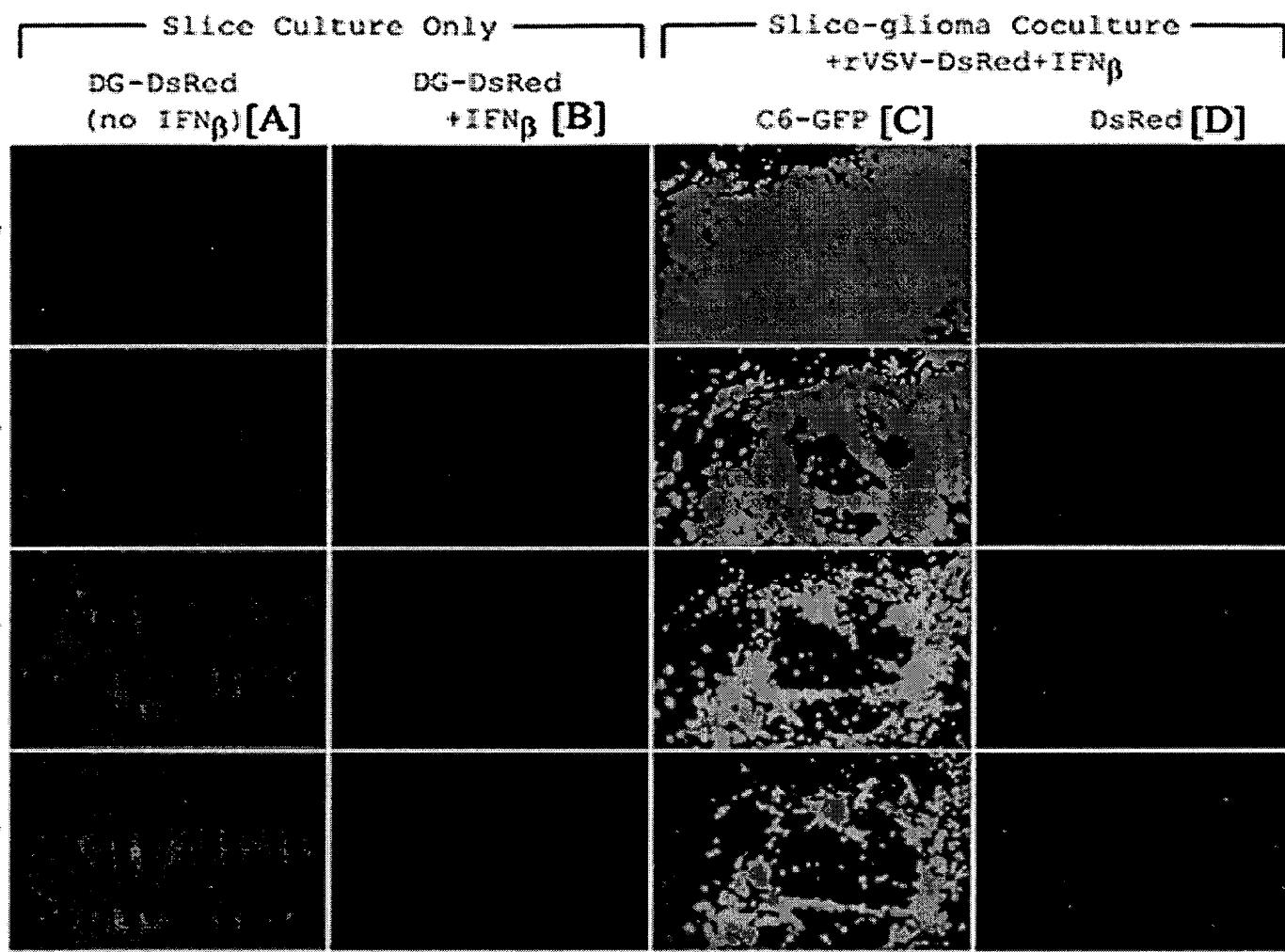


Figure 39

MAP2 Immunohistochemistry

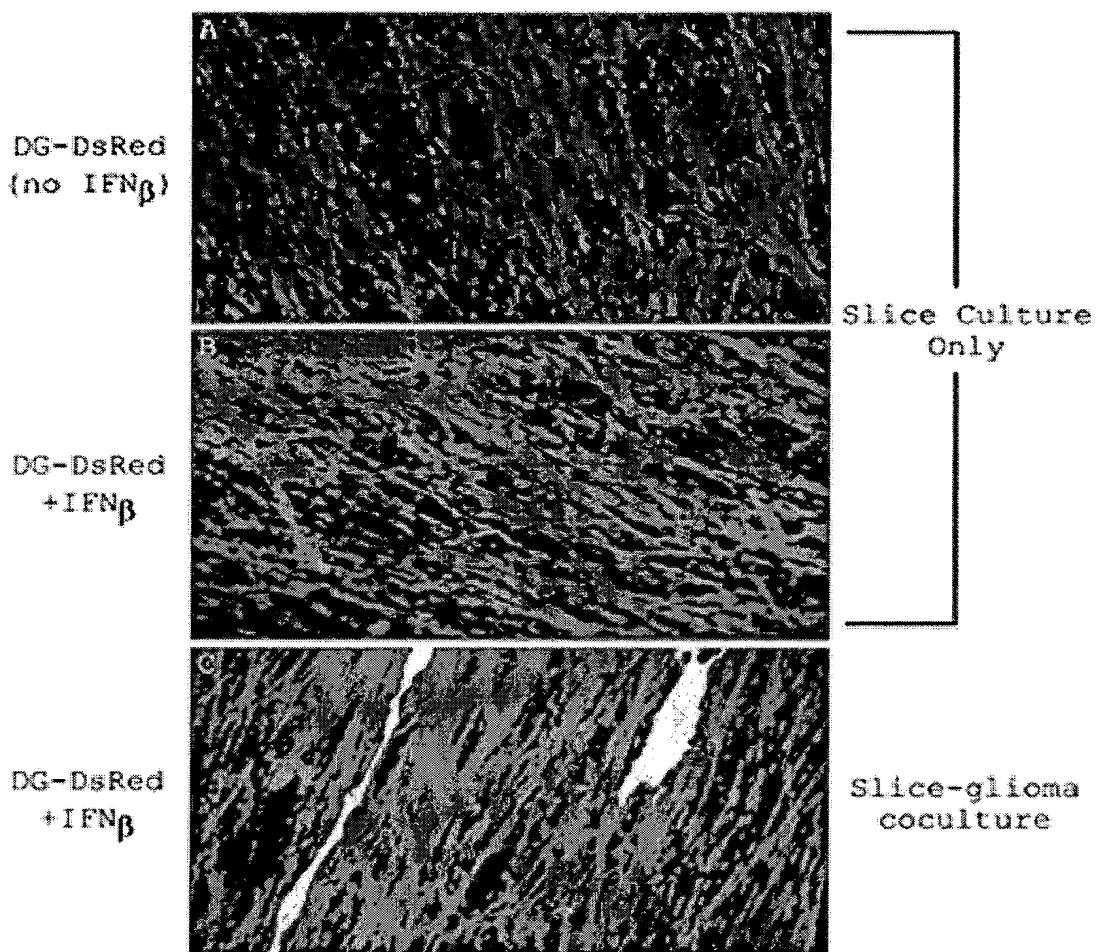


Figure 40

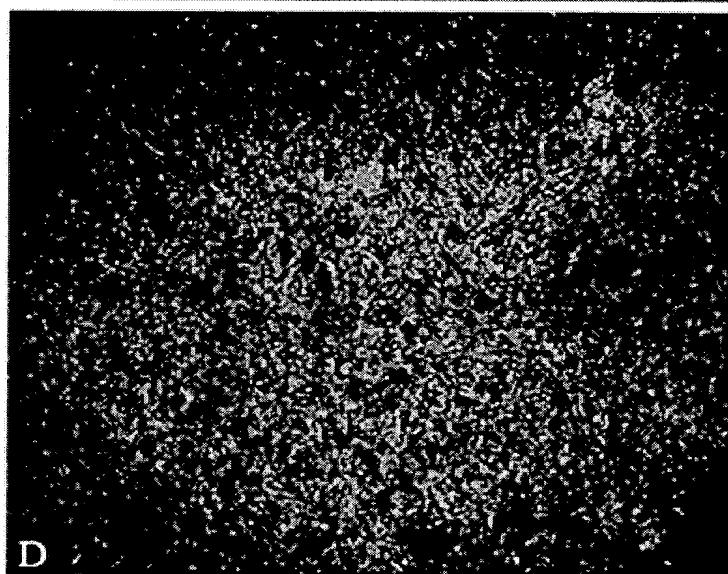
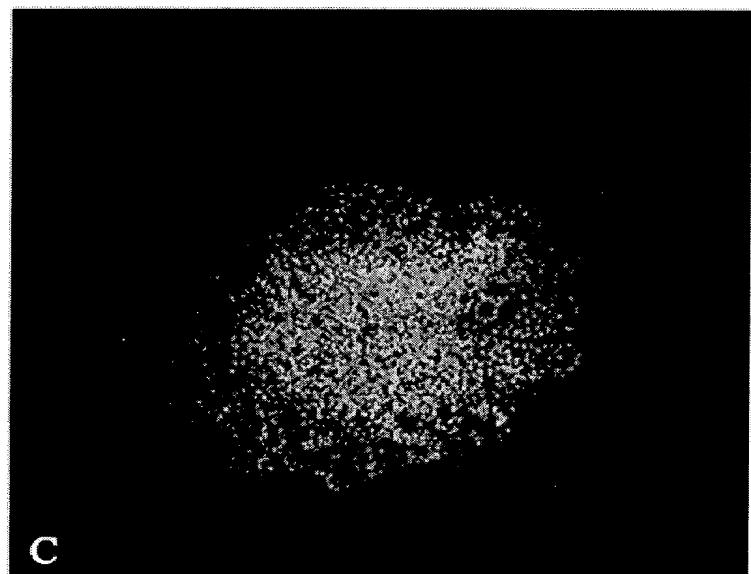
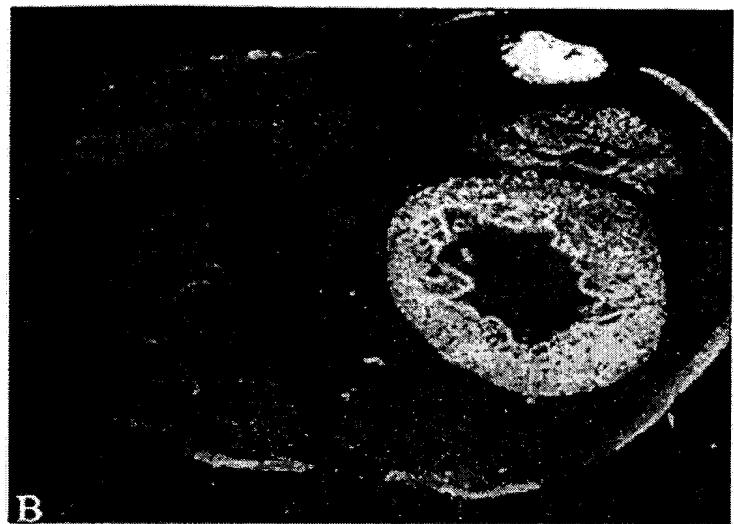
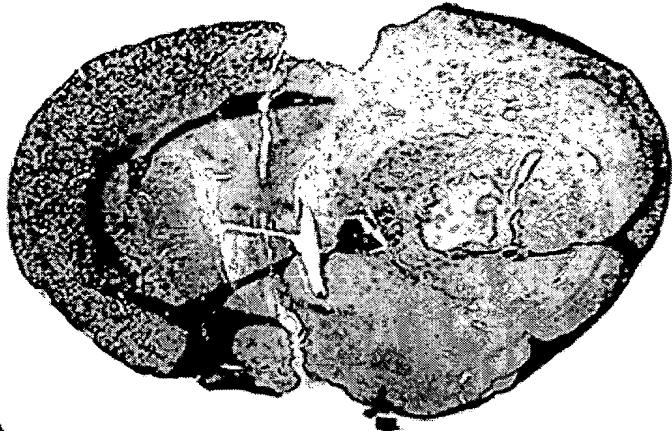
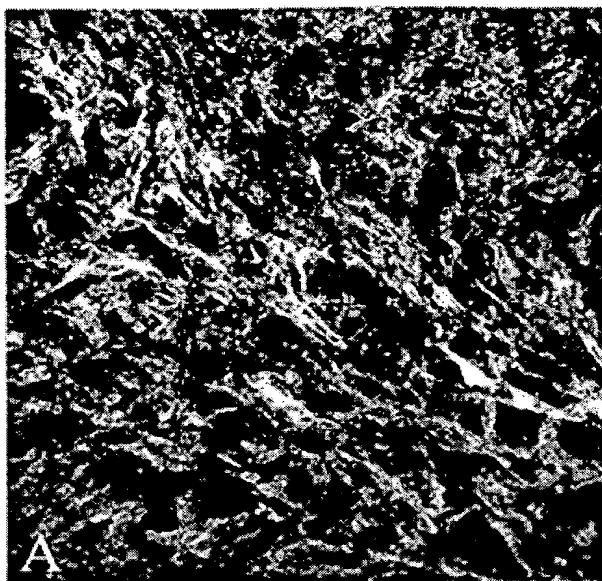
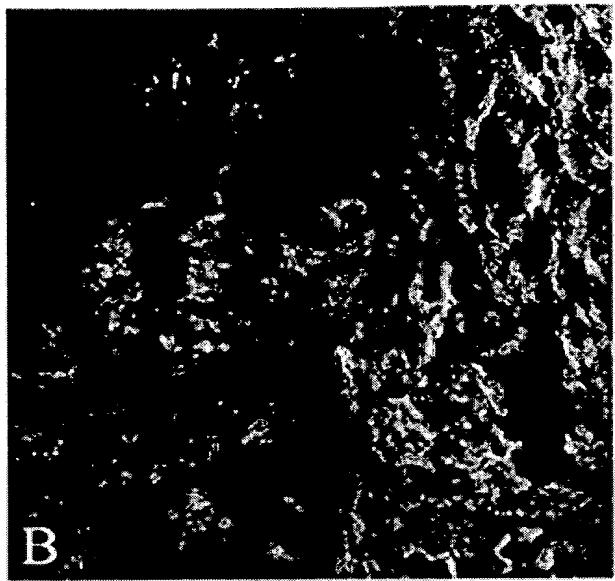


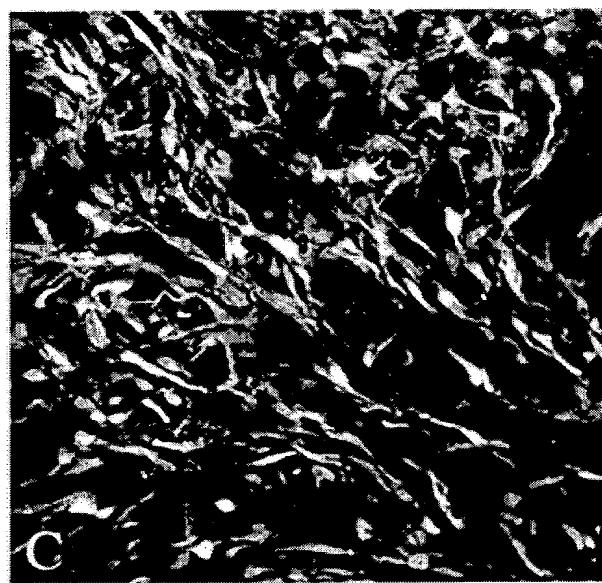
Figure 41



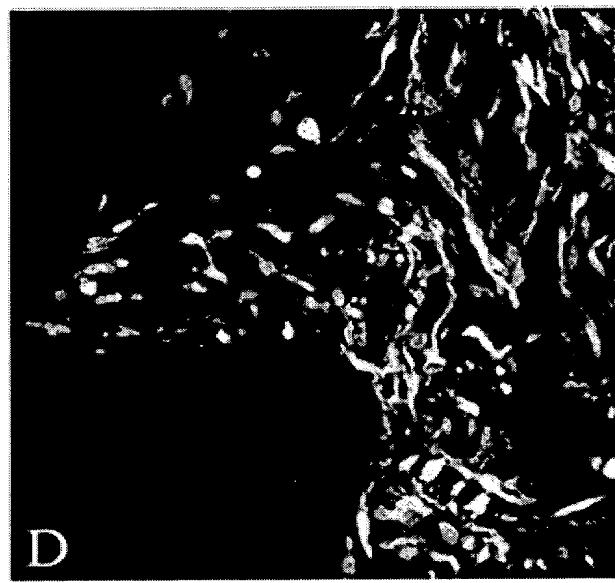
A



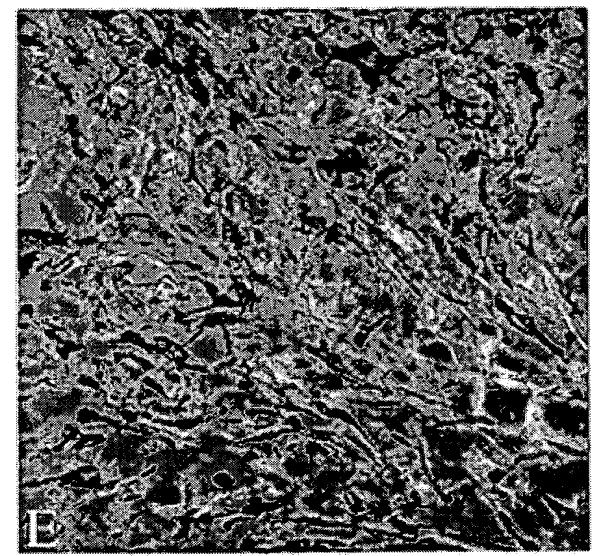
B



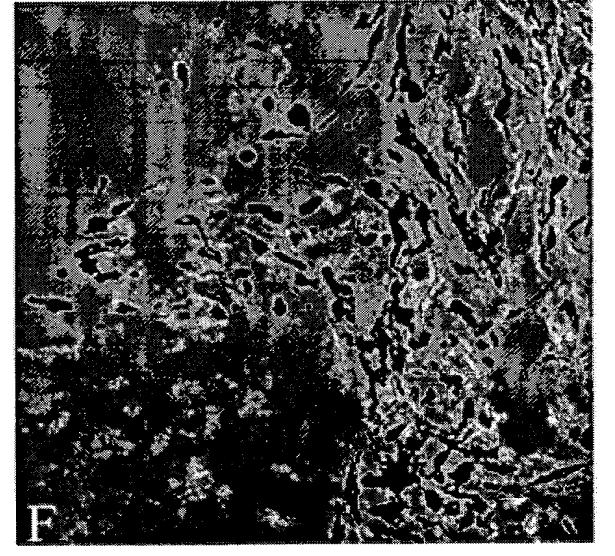
C



D



E



F

Figure 42